TO STUDY CALCINEURIN INHIBITOR SPARING STRATEGIES IN PATIENTS WITH STEROID RESISTANT AND STEROID DEPENDENT NEPHROTIC SYNDROME

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Dedicated

to

my family

&

Teachers

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Dr. Mahak

LIST OF ABBREVIATIONS

- AKI Acute Kidney Injury
- BMI Body Mass Index
- CKD Chronic Kidney Disease
- CNI Calcineurin inhibitor
- CR Complete Remission
- CsA Cyclosporine A
- eGFR Estimated Glomerular Filtration Rate
- FRNS Frequently Relapsing Nephrotic Syndrome
- FSGS Focal Segmental Glomerulosclerosis
- INS Idiopathic Nephrotic Syndrome
- IPNA International Paediatrics Nephrology Association
- IQR Interquartile Range
- ISKDC International Study of Kidney Disease in Children
- KDIGO Kidney Disease Improving Global Outcomes
- LEV Levamisole
- MCD Minimal Change Disease
- MMF Mycophenolate Mofetil
- MPA Mycophenolic Acid
- MPS Methylprednisone
- NS Nephrotic Syndrome
- OPD Outpatient Department
- OR Odds Ratio
- PDN Prednisolone
- PR Partial Remission
- RCT Randomised Controlled Trial
- RR Relative Risk
- SPSS Statistical Package for the Social Science
- SRNS Steroid Resistant Nephrotic Syndrome
- SSNS Steroid Dependent Nephrotic Syndrome
- TAC Tacrolimus
- Up/Uc Urine protein to creatinine ratio

ABSTRACT

INTRODUCTION: Steroid resistant nephrotic syndrome (SRNS) and steroid dependent nephrotic syndrome(SDNS), while uncommon, are associated with significant morbidity in children. Calcineurin inhibitors (CNIs) remain the first line recommended therapy for children with non-genetic forms of SRNS and have been used successfully in children with SDNS as well. Although efficacious, long term use is hindered by their adverse effects. Thus, alternative therapies are needed for maintaining sustained remission in such patients on CNI.

AIMS and OBJECTIVES: This study aimed to study various CNI sparing treatment strategies being used in SRNS and SDNS to maintain sustained remission.

METHODS: The study was conducted retrospectively and prospectively over a period of 1 year to include patients below 18 years of age, with SRNS or SDNS; who had received a minimum of 12 months therapy with any of the calcineurin inhibitors (i.e. cyclosporine or tacrolimus) and were either in complete or partial remission at the time of enrolment. Patients with secondary or genetic forms of nephrotic syndrome were excluded. Data abstracted included demographics, histopathology on renal biopsy, prior and concomitant use of other immunosuppressive agents, and serial clinical/laboratory data. Primary outcome measure was incidence of sustained remission (absence of relapse and steroid free period in six months period). The secondary objectives were to observe the incidence of relapses and side effects with the strategy used.

RESULTS: Our study included 18 children with SDNS and 11 with SRNS. The majority were male. The most common CNI sparing agent used was rituximab in 19 (65%) followed by cyclophosphamide in 6 (20.7%), methotrexate in 2 (6.9%) while mycophenolate was used in only one patient of SDNS. 25 of these patients were followed up for 12 months. Sustained remission was seen in 9 out of 25 children (36%) and 66.6% of these patients were given Rituximab (3 infusions of 375 mg/m^2 in majority) as CNI sparing agent. The mean time to first relapse was 5.6 months and there was a reduction in incidence relapse rate from 1.55 to 0.82 relapses/patient year. At twelve months follow up, all patients (n=17) on rituximab therapy were in remission (88.8% CR, 11.7% PR); 3 (75%) patients on oral cyclophosphamide were in complete remission while one was in partial remission; both patients on methotrexate were in complete remission (but incidence relapse rate was high;1.5); the only patient of SDNS who was given mycophenolate was in complete remission at 12 months with no relapse. Only one patient was in non remission at 1 year (SDNS patient; given cyclophosphamide). There was a significant improvement in eGFR after switching therapy from CNI to other less nephrotoxic agents (p=0.03). No major side effects were seen with any of the therapy.

CONCLUSION: Rituximab appears to be an effective option for maintaining remission in patients of SDNS and SRNS who have already received CNI for a long term; oral cyclophosphamide showed mixed results; MMF seems to be a safer and effective option in SRNS for maintaining remission; though sample size was small.

KEYWORDS: Steroid resistant nephrotic syndrome, steroid dependent nephrotic syndrome, Rituximab, Cyclophosphamide, Methotrexate, mycophenolate mofetil, remission, relapse.

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KEY TO MASTER CHART

- BMI Body Mass Index
- CNI Calcineurin Inhibitor
- CR Complete Remission
- creat Creatinine
- eGFR estimated Glomerular Filtration Rate
- F Female
- FRNS Frequently Relapsing Nephrotic Syndrome
- M Male
- PR Partial Remission
- SDNS Steroid Dependent Nephrotic Syndrome
- SRNS Steroid Resistant Nephrotic Syndrome
- TLC Total Leucocyte Count
- UTI Urinary Tract Infection
- uPCR urine Protein to Creatinine ratio

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INTRODUCTION

Idiopathic nephrotic syndrome (INS) is a common cause of chronic kidney disease (CKD) in children. The estimated incidence is 2.92 (range 1.15-16.9) per 100,000 children/year.[1] It is defined by the presence of proteinuria in nephrotic range; ≥3+ by dipstick, or proteinuria ≥ 40 mg/m^2/hr (>1000 mg/m^2/day), Up/Uc ≥2 mg/mg; hypoalbuminemia (<3 g/dl); and edema.[2]

Childhood NS was linked to a high mortality secondary to various causes like acute kidney injury (AKI), chronic kidney disease (CKD), systemic infections, and thromboembolic events prior to the discovery of immunosuppressive agents. In 1950s, glucocorticoids were shown to be an effective treatment to induce remission. Since then, the overall prognosis of patients with NS has improved dramatically.

In the majority, approximately 85% of cases, complete remission of proteinuria is seen within 4-6 weeks with daily prednisolone/prednisone (PDN) treatment at standard doses. These patients are labelled as steroid sensitive nephrotic syndrome (SSNS).[3] Those patients who do not achieve remission after 4-6 weeks of therapy with prednisone are presumed to have steroid resistance and labelled as steroid resistant nephrotic syndrome (SRNS).[4]

It is observed that among the patients with SSNS, 70-80% have atleast one relapse during follow up. About 50% of these have ≥ 4 relapses/year) i.e. frequent or become steroid-dependent (two consecutive relapses on steroid therapy or within 14 days of stopping steroids); referred to as having steroid dependent nephrotic syndrome (SDNS).[5]

The SSNS with onset in childhood may resolve spontaneously during puberty; however, 10-30 % of these may still continue to have a relapsing course into young adulthood.[6] Management of frequently relapsing SSNS and SDNS is quite challenging, because prolonged or frequent use of high dose steroids leads to steroid toxicity and a reduction in overall life quality. IPNA guidelines recommend that steroid sparing agents be used in children with SDNS (grade B, strong

recommendation). The immunosuppressive drugs used as steroid sparing agents are levamisole, cyclophosphamide, mycophenolate mofetil (MMF), and calcineurin inhibitors (CNIs). Rituximab is recommended in children having SDNS who do not show any response to treatment with any one of the steroid sparing agents at adequate dose.[7]

The treatment of patients with steroid resistance has many challenges as well; due to variable response to immunosuppression therapy, due to disease related adverse effects, risk of progression to kidney failure, and likelihood of post-transplant recurrence.[4] The intention to treat with any immunosuppressive agent remains the same i.e. to induce remission of proteinuria and lead to favourable long term outcomes.

Calcineurin inhibitors (CNI) have been recommended in the guidelines as the first line therapeutic agents for patients who have initial or late steroid resistance [5], however, the optimal duration of CNI therapy varies in different guidelines. KDIGO and IPNA guidelines recommend giving CNI for a minimum of 12 months if patients show partial remission. And in patients who show complete remission, discontinuation of CNIs after 12-24 months and consideration of starting MMF for maintaining remission has been suggested.[2]

High-dose CNI, when used for a prolonged duration can result in undesirable side effects (e.g. nephrotoxicity). To overcome this, alternative or additive drugs have been sought; including iv or oral cyclophosphamide, rituximab, MMF and monoclonal antibodies such as rituximab, ofatumumab. Despite various studies and trials, a consensus regarding the effectiveness and safety profile of available pharmacological options during long term follow up, to maintain remission, is lacking.

Mycophenolate mofetil (MMF) is an ester prodrug of mycophenolic acid (MPA). It is thought of as a promising therapeutic option for maintaining remission in patients who have SRNS and SDNS. It has been reported in case series that therapy with MMF is effective and safe in decreasing the relapse frequency and the cumulative steroid dose in steroid-dependent and resistant nephrotic syndrome.[8]

Cyclophosphamide (CP) is an alkylating agent that was introduced in 1967 as a part of nephrotic syndrome treatment protocols. It was the first cytotoxic drug shown to effectively reduce steroid requirement in children with this condition. Data regarding children of Indian descent with SRNS treated with IV cyclophosphamide indicate that a small number of patients can achieve complete remission (CR), whether it is used as first line therapy or following a high dose intravenous steroid trial. Thus, it is recommended as the first line therapeutic option for children with SRNS who are not responding to or cannot be treated with calcineurin inhibitors.[9,10]

Levamisole, an anti helminthic drug, has immunomodulatory properties as well. It has been effectively used at a dosage of 2.5 mg/kg/day alongside alternate day prednisolone for the treatment of SDNS. There is sufficient evidence that addition of levamisole can contribute towards a reduction of maintenance dose of corticosteroids and result in a prolonged remission period.[11]

Rituximab is a chimeric anti-CD20 antibody (monoclonal) that was previously used for managing B-cell non-Hodgkin's lymphoma. Many observational studies and RCTs have shown the successful use of rituximab in difficult to treat cases of nephrotic syndrome.[12] Recent studies show that Rituximab may be used for maintaining remission of INS as effectively as standard therapy including prednisone and calcineurin inhibitors.[13]

The aim of this study was to observe the various CNI sparing treatment strategies being used in a tertiary care centre to maintain sustained remission in patients with SRNS and SDNS who have received CNI for atleast twelve months and are in remission (either partial or complete) with the same.

AIMS AND OBJECTIVES

AIM: To study the various CNI sparing treatment strategies in SRNS and SDNS after atleast one year of therapy with calcineurin inhibitors

PRIMARY OBJECTIVE: To observe the incidence of sustained remission after change in therapy

SECONDARY OBJECTIVES:

1. To observe the incidence of relapses prior to and after the change in therapy
2. To observe the side effect profile prior to and after change in therapy including renal dysfunction (eGFR)

REVIEW OF LITERATURE

Management of both SDNS and SRNS presents significant challenges for the treating clinicians. They often face therapeutic dilemmas while treating children with FRNS/SDNS and steroid-resistant nephrotic syndrome ; requiring careful consideration of treatment options.

Management options for SDNS:

Children with SDNS often need treatment with other immunosuppressive, steroid sparing agents like alkylating agents (cyclophosphamide), levamisole, rituximab, mycophenolate mofetil (MMF), and CNIs (cyclosporine, tacrolimus) for mitigating the glucocorticoid-related adverse effects and for improving the clinical course.

Studies have earlier shown that second-line therapies show efficacy in reducing the number of relapses in children with FRNS or SDNS compared to either glucocorticoids alone or placebo. But there are few RCTs that show direct comparison of the efficacy and safety of various interventions. Therefore, as robust evidence is lacking , the selection of steroid sparing treatment is based on patient's age, features of steroid toxicity, adherence to treatment, risk of side effects, patient preferences, and the availability of medications.

Levamisole

Among the various agents used as steroid sparing agents in SDNS and FRNS, levamisole (LEV) has been shown to be the least toxic and the least expensive option. The relapse rate as well as cumulative steroid dose with levamisole had been reported to be significantly reduced in the previous studies by Sumegi and Madani et al.[11]

Gruppen (2018) et al compared LEV therapy to placebo in a study involving 99 children with FRNS or SDNS in an international, multicentre, double blind RCT using placeo as control. They reassessed the use of levamisole in preventing relapses in children with SSNS. In their trial, a significant reduction in the number of relapses at 12 months with a relative risk (RR) of 0.77 for relapses in the LEV group (95% CI: 0.61 to 0.97) was noted. Consequently, 26%

of children in the LEV group maintained remission at 12 months, compared to only 6% in the placebo group. Thus, they showed that using levamisole resulted in an increase in time to relapse and also prevented recurrence compared to prednisolone alone.[14]

Mycophenolate mofetil

MMF, the prodrug of MPA inhibits the proliferation of B- and T-lymphocytes, thereby suppressing the immune system. It acts as a potent, reversible and non competitive inhibitor of inosine monophosphate dehydrogenase (IMPDH), the most important enzyme involved in the de novo synthesis of purines in proliferating T , B lymphocytes. It has a beneficial side effect profile in contrast to CNIs, specifically lack of renal toxicity and is studied for steroid sparing in many recent studies. The correlation between MMF dose and mycophenolic acid (MPA) exposure is poorly established leading to significant inter- and intra-patient variability in drug pharmacokinetics.

Role of MMF in SDNS

Many studies have reflected the efficacy of MMF in comparison to prednisolone alone for maintenance of remission in children with FRNS or SDNS.[15]

Experience from uncontrolled studies suggests that mycophenolate mofetil (MMF) may be more effective in young children and potentially superior to levamisole in managing steroid dependence.

No major RCTs have so far examined the efficacy of MMF alone compared with no treatment or glucocorticoids alone in patients with FRNS or SDNS.

In 2019, Saravanakumar Karunamoorthy et al. investigated the use of MMF in children, adolescents with steroid-dependent nephrotic syndrome (SDNS). The study involved 87 patients, with a male-to-female ratio of 2:1. The median age at diagnosis of NS was 3 years (95% confidence interval [CI]: 1–8 years). Intravenous cyclophosphamide was administered as the first-line treatment to 82 patients; of these, 24 experienced persistent proteinuria, while 58 achieved remission for a median duration of 6 months. The median duration of MMF treatment was 2 years and 6 months (95% CI: 1 year

and 3 months to 4 years and 6 months). Among the participants, 72 patients (83%) were sensitive to MMF, leading to a decline in the mean prednisolone dose from 1.28 to 0.35 mg/kg ($p<0.05$). The study concluded that continuous MMF therapy showed remission in 83% of the patients, and it was well tolerated. However, discontinuing MMF resulted in a 100% relapse rate.[16]

However, MMF has demonstrated lower efficacy compared to calcineurin inhibitors (CNI) in various studies. Dorresteijn et al. enrolled 24 children over a 12-month period and found that rate of relapses was higher in patients given MMF instead of those on cyclosporine A (CsA), with a p-value of 0.08 after twelve months of treatment.[17]

The higher efficacy of cyclosporine A than MMF was also shown in a randomised multicentre, open label controlled trial conducted by the German Society for Pediatric Nephrology. They enrolled 60 patients with frequently relapsing SSNS. The primary endpoint studied was the relapse frequency. It was seen that the incidence relapse rate was more (statistically significant) with MMF as compared to cyclosporine A ($p=0.03$). Patients were in sustained remission without relapse for significantly longer period in patients on CsA than on MMF during the first 12 months of therapy ($p<0.05$) and not during the second year ($p=0.36$).Thus, this study showed the inferiority of MMF than CsA for prevention of relapses in pediatric patients with FR-SSNS.[18]

Alkylating agents

Cyclophosphamide and chlorambucil are the two alkylating agents extensively studied in the management of nephrotic syndrome. Cyclophosphamide is being used in NS since 1963. Barratt et al, in 1970, conducted the first controlled trial of cyclophosphamide in FRNS and demonstrated the efficacy of cyclophosphamide in this population.[19]

A retrospective study by Azib S et al was done in 2011, in which they studied data from 90 patients with SDNS; all of whom were given a single course of oral cyclophosphamide (2 mg/kg/day for 10 to 12 weeks). Patients were followed up for a median period of 5.5 years, sustained remission was observed in 57% patients at 1 year, 42% at 2

years and 31% at 5 years. There was a significant decrease in the median dose of prednisolone use between cyclophosphamide initiation and first relapse (22.1 mg/kg/day versus 4.9 mg/kg/day, $p < 0.001$). Around 60% of all patients remained free of any further immunosuppressive agent. Thus, a short course of cyclophosphamide was shown as an effective second line therapy in SDNS patients.[20]

Around 35 studies on the effectiveness of cyclophosphamide in SDNS have already been done and these were collected in a meta-analysis that comprised of 1504 children. It was seen that, as the cumulative dosage of chlorambucil and cyclophosphamide increased, the relapse free survival also increased. A safe threshold for a cumulative cyclophosphamide dose could not be found in males. They recommended a dose of 2-3 mg/kg body weight for 8-12 weeks as the standard dose for oral cyclophosphamide.[21]

Calcineurin Inhibitors:

Calcineurin inhibitors (CNIs) inhibit T-cell activation, the possible mechanism of their effect in nephrotic syndrome treatment. Cyclosporine and tacrolimus are the main CNIs used. They were effective for maintaining remission in children with FRNS and SDNS in various studies.

The effectiveness of cyclosporin A (CsA) in the reduction of relapses in patients with SDNS and in inducing remissions in an important number of patients with non genetic forms of SRNS has been well elucidated in studies.[22]

A randomised controlled trial was performed in Japan in 1994 that included 108 children with FRNS/SDNS showing that the risk of relapse decreased significantly with CsA when compared to placebo [(relapse rate 0.55 (95% CI 0.37–0.82)].[23] Observational studies too have shown decreased relapse rates with CsA compared with PDN.

Tacrolimus is the preferred agent over cyclosporine nowadays in many centres because of its relatively better efficacy and less systemic side effects, particularly nephrotoxicity. This excludes children who cannot swallow tablets, as well as patients who suffer from seizures or those at a risk of diabetes. The doses of calcineurin inhibitors (CNIs) are

adjusted to get the recommended trough levels, while also considering potential interactions with other medications.

Rituximab in SDNS:

Rituximab is a chimeric antibody (monoclonal) that targets CD20, an antigen present at various stages of development of B cells. Its binding to CD20 leads to rapid depletion of the populations of B cells and has been utilized in the treatment of several autoimmune disorders.[24] The exact mechanisms leading to primary NS still remain unclear and continuous research is going on in this field. According to recent evidence, B cells do have an important role in the development of INS and thus are being studied as a target for therapy.

The earliest evidence of benefits of rituximab in INS was in case reports published in the early 2000's. It was initially limited to case reports and small case series mainly consisting of patients with SDNS or FRNS. These studies demonstrated significant response to rituximab, often resulting in prolonged remission and allowing patients to be weaned off other immunosuppressive agents.

It is being more and more used for the management of INS as seen in the last decade, and has shown variable responses in children of SDNS, FRNS or SRNS.[25,26]

In a randomised trial conducted by Ravani et al, 30 children with SDNS, who required steroids in high dose for maintaining remission, were enrolled. These children were not given any of the calcineurin inhibitors previously and showed a non-complicated course of SDNS. The minimum follow up period was 1 year. Children in the treatment group (n = 15) were given one dose of rituximab (375 mg/m2). After 3 months, the proteinuria was only modestly reduced (42% lower than the control group, $P = 0.58$), but the patients required significantly lesser doses of steroids at 3 months ($P < 0.001$) and further, a substantially reduced risk of relapses at 6 months was seen. This reduced risk continued to be there at 1 and even 2 years after treatment ($P < 0.01$). After the therapy with Rituximab, the median time to relapse was 18 months in this study.[27]

Liu et al in 2021 conducted a meta analysis on the effectiveness and safety profile of Rituximab for Childhood onset SDNS. This meta analysis was done on six RCTs and also included one comparative control study. A total of 446 patients were enrolled. 231 patients out of these were in the rituximab arm and 215 were in the control arm . The data analysis showed that a significantly improved rate of CR occurred in the rituximab group in contrast to the control group (OR =5.21; 95% CI, 3.1-8.5; p<0.0001). Also, there was a statistically significant increase in the level of serum albumin after therapy with Rituximab (MD= 0.88, p = 0.001) and eGFR (MD= 6.43, CI 2.6-10.1, p = 0.0008). but there was insignificant change in the level of serum creatinine and proteinuria.[28]

Management options in SRNS:

Managing the patients with SRNS is equally challenging. They report a poor quality of life and experience morbidity because of infectious and non infectious illnesses and also due to side effects of various immunosuppressive and other medications used for their treatment. The chance of renal dysfunction is significantly greater for patients who do not to achieve remission with any therapy.[4]

The most frequently used second-line agents used in SRNS patients include cyclosporine, tacrolimus, high-dose i.v. methylprednisolone, and MMF. These drugs have been variably used in patients with SRNS, though the response rate is not as good as patients with SDNS, FRNS.

Role of MMF in SRNS

MMF monotherapy has been assessed in uncontrolled studies involving patients with SRNS, with varying percentages of patients who achieve remission with treatment.

A randomized multicentre trial (funded by National Institutes of Health) was conducted by Gipson in 2011. In this trial, they compared the effectiveness of a 12-month course of cyclosporine with a combination of MMF and oral steroid (dexamethasone) in patients with SRNS,

biopsy proven FSGS. They reported remission (partial or complete) in 46% of patients who were on cyclosporine and 33% of patients on MMF + dexamethasone after 12 months treatment. The rates of remission of proteinuria were similar in the two groups.[29]

Li Z et al enrolled 24 children (18 boys and six girls) under 2 years of age, with steroid-resistant idiopathic nephrotic syndrome (SRINS). All patients were subjected to a course of prednisone 2 mg/kg per day. After eight weeks of prednisolone treatment, MMF was started at 25-30 mg/kg/day dose and given for a duration of 6-12 months. Prednisone dose was slowly tapered. With the use of MMF, they reported a decline in proteinuria and serum cholesterol and an increase in serum albumin in a time-dependent manner. It was demonstrated in their study that MMF could reduce proteinuria in SRNS patients who were below 2 years and it was suggested that MMF therapy could be an effective treatment option in this age group.[30]

In an open-label, one-to-one randomised, controlled trial, Sinha et al. investigated the impact of transitioning from tacrolimus medication to MMF on the disease course in children (aged one to eighteen), who were recently diagnosed with steroid-resistant nephrotic syndrome, at an Indian referral centre. Patients with complete or partial remission after therapy with tacrolimus for six months were randomised so that 29 received MMF and 31 received tacrolimus plus tapering steroid on alternate days for twelve months. Intention-to-treat analyses revealed that the MMF group had a substantially lower number of patients (44.8%) with a favourable outcome (sustained remission, uncommon relapses) after one year in comparison to the tacrolimus group (90.3%). Patients given MMF experienced a considerably higher frequency of relapses than tacrolimus group (mean difference: 1.05 relapses per person-year). Patients on MMF had significantly (71%) lower chances of a favourable result and significantly higher chances of treatment failure (recurrences, steroid resistance) in comparison to those on tacrolimus.

For children with SRNS, switching from tacrolimus to MMF after six months tacrolimus treatment was linked to an increased risk of recurrent relapses or resistance.[31]

The IPNA clinical guidelines on SRNS suggest using MMF in SRNS patients and those with eGFR < 30 ml/min/1.73 m^2 due to the higher risk of nephrotoxicity with CNI and for maintenance of remission in SRNS patients who are already on CNI.[2]

Cyclophosphamide in SRNS

Cyclophosphamide in SRNS was first studied in two trials , the Tarshish trial and the ISKDC trial. These trials compared oral cyclophosphamide therapy plus corticosteroids to corticosteroids alone. There was not a significant difference in achieving a complete remission in both the groups in both the trials.[32]

However, the role of iv cyclophosphamide administered as monthly doses for six months in combination with tapering prednisolone doses on alternate days was earlier studied in a number of case series. This data shows that such therapy leads to remission in 40-65% of patients.

In study by Bajpai et al, the effect of treatment with iv cyclophosphamide in patients with SRNS was studied. 24 patients were enrolled into the study and iv cyclophosphamide was given once a month for 6 months combined with alternate day steroid. Complete proteinuria remission was observed in 29.2% of patients and was sustained on long term follow up in 20.8%. They concluded that iv cyclophosphamide treatment could be beneficial to patients with late steroid resistance. [33] Thus, data on children of Indian ascent given iv cyclophosphamide have shown response in the form of CR in a small subset of patients, either when iv cyclophosphamide was used as first line therapy or after failure of a trial of high dose iv steroids. Thus, iv cyclophosphamide has been weakly recommended for SRNS patients who are unable to be treated with CNIs.[2]

Another study in North American children with difficult to treat SRNS demonstrated that iv cyclophosphamide could be a useful strategy for the induction of remission in a subset of children with MCD on biopsy.

Fifty-percent of the children treated with IV Cyclophosphamide went into complete remission and it was sustained over a considerable period of time, with preserved renal function. This study showed favourable response with Cyclophosphamide in around 50% patients with difficult to treat nephrotic syndrome.[34]

Consensus guidelines on management of SRNS [5] do suggest treatment with iv cyclophosphamide for patients in whom CNI cannot be given, either due to the higher cost or adverse effects.

Calcineurin inhibitors in SRNS:

CNIs have shown to be more effective than cyclophosphamide and MMF in SRNS patients. In various studies, use of CNIs was associated with more remission rates as compared to no treatment in SRNS.[35]

The use of CNI as first-line agents in SRNS patients in childhood was assessed in various RCTs that compared the effectiveness of cyclosporine (CsA) with either placebo [36], intravenous methylprednisolone [37] or tacrolimus (TAC) [38] and CsA or TAC with intravenous Cyclophosphamide [39] on the outcome of "number with complete or partial remission".

In a multicentre, randomised controlled trial conducted in 2012 by Gulati et al, 131 consecutive paediatric patients were randomly given IV cyclophosphamide infusions every six months or tacrolimus for six months.

Prednisolone was administered in equal doses to both arms on alternate days. Patients in tacrolimus group showed a much greater rate of complete remission (52.4%) compared to cyclophosphamide (14.8%). Compared to cyclophosphamide, tacrolimus significantly increased the secondary outcome of prolonged remission or steroid-sensitive recurrence of nephrotic syndrome at 12 months.[39]

Li et al conducted a meta analysis on the study of effect and safety profile of different immunosuppressive medications for SRNS in children. Both tacrolimus and cyclosporine showed decreased levels of plasma cholesterol, serum creatinine, and proteinuria when compared to the placebo/no treatment group, iv or oral cyclophosphamide,

azathioprine, chlorambucil. Two studies comparing CsA to TAC were included into the meta-analysis to assess the efficacy of CsA in SRNS patients. There were no significant group differences in complete remission (CR: OR = 1.71, 95%CI: 0.58–5.04, *P* = 0.33).[40]

Due to their nephrotoxic effects, CNI treatment is avoided for patients with decreased eGFR, acute kidney injury and/or uncontrolled hypertension. Most patients of SRNS who respond to CNI do so within first 6 months of treatment.[39] Patients classified as CNI resistant are those with SRNS who do not show even a partial response to CNI after 6 months. Individuals who, after 12 cumulative months of therapy, show no improvement with CNI plus another medication that is mechanistically different are classified as multi-drug resistant.

Rituximab in SRNS:

Rituximab, like other drugs used for NS, is less successful in SRNS patients than in SDNS/ FRNS patients.

A few years back, Magnasco carried out a RCT including 31 SRNS children. Of all the participants, 16 received CNIs plus prednisolone plus two rituximab infusions, whereas the remaining 15 received CNIs plus prednisolone alone. Unfortunately, neither total nor partial remission was attained by any study participant, and there was no visible variation in the level of proteinuria between the rituximab and control groups.[41]

Sinha et al. did a comprehensive retrospective analysis in which they observed that 58 patients with steroid- and CNI-resistant nephrotic syndrome responded well to rituximab (2–4 doses), (a few of them did not respond to cyclophosphamide either), while 17 patients had positive reactions [PR (17.2%) in 10 and CR in 7 patients (12%)].[42]

Kamei et al studied the effect of rituximab treatment in combination with methylprednisolone pulse therapy and other immunosuppressants for childhood onset SRNS. They enrolled ten patients with calcineurin inhibitor and MPS resistant SRNS and managed them with rituximab (1-4 doses; 375 mg/m^2 each) followed by methylprednisolone once every 2-4 weeks until CR was achieved. They concluded that refractory

SRNS may be treated with rituximab in addition to immunosuppressive medications and traditional MPS.[43]

Optimal duration and Nephrotoxicity of CNI:

In the absence of studies that determine the appropriate duration of CNI therapy in SRNS patients with remission, KDIGO and IPNA recommend therapy for at least 12 months, while ISPN advises therapy for 2 years or longer. Treatment should be monitored closely for adverse effects. [2, 35]

10% to 25% of patients on long-term CNI treatment experience nephrotoxicity.[44] Risk factors for CNI induced nephrotoxicity include initial resistance to CNI, CNI dosage, length of severe proteinuria, and hypertension during treatment. Nephrotoxicity caused by cyclosporine is primarily caused by chronic ischaemic injury to the kidney. This insult leads to tubulointerstitial alterations, such as striped interstitial fibrosis, tubular vacuolization, and atrophy, and arteriolar hyalination. [45]

Similar to CsA, tacrolimus has been reported to be associated with renal interstitial fibrosis. A substantial correlation between elevated TAC trough levels and presence of renal interstitial fibrosis has been reported in the study by Morgan C et al.[46]

It is essential to transition to less toxic drugs in order to maintain the CNI-induced remission due to adverse pharmacological effects, which include glucose intolerance and acute and chronic nephrotoxicity.

Because mycophenolate mofetil (MMF), the prodrug of mycophenolic acid, has no reported adverse effects on the kidneys, haemodynamics, or metabolism, it could be a favourable choice for the treatment of difficult cases of nephrotic syndrome.[47]

Using a standard protocol that included MMF (40 mg/kg/day) in addition to prednisolone and cyclosporine for maintenance medication and methylprednisone pulse therapy for patients who continued to experience relapses, Hibino et al. examined the three-year outcomes of 91 patients with SDNS or SRNS. MMF was effective in reducing the dose of steroids in maintaining emission in patients with SDNS. Individuals with SRNS were able to achieve remission rates of 67%

(8/12) in FSGS and 100% (6/6) in MCD. Consequently, they discovered that CsA therapy and the combination of CsA and MMF were effective for treating SDNS and inducing remission in SRNS. [48]

Gellerman et al. conducted a retrospective analysis involving 23 patients who had achieved CR with cyclosporine medication. The study aimed to wean CsA and sustain remission using MMF monotherapy. This protocol maintained remission in all patients. Throughout a 7-year follow-up period, 5 out of 23 patients (21%) had 10 relapses in total, all of which were treated with therapy. At the conclusion of their investigation, all 23 patients were in remission, with MMF alone (n = 13; 57%), or with a combined therapy of CsA and MMF (n = 2, 9%). In seven individuals (30%), therapy could be stopped permanently.[49]

In certain individuals with CNI dependence and/or toxicity, retrospective case records demonstrate that MMF allows remission without the need for corticosteroids or CNI.

Fujinaga S et al. examined 12 children having INS who needed long-term CsA for a minimum of 6 months in a prospective study on the use of MMF in patients with cyclosporine-dependent nephrotic syndrome. With a decrease in mean dosage of CsA from 3.5±1.3 to 1.5±2.4 mg/kg/day ($p<0.01$) and mean prednisolone dose from 0.29±0.16 to 0.21±0.11 mg/kg/day ($p<0.05$), therapy with MMF led CNI and/or prednisolone (PSL) sparing. Thus, MMF therapy significantly reduced the risk of relapse in children with CsA-dependent NS and spared CsA and/or steroids in this study.[50]

Out of the four case series that on patients with CNI-refractory disease, it was demonstrated that a combination of CNI and MMF may produce remission in 67.3% of the patients, without substantial side effects.[51]

Children with INS who were having sustained remission for at least six months and depended on prednisone and CNI for at least 12 months were the study participants of long-term follow-up data published by Ravani et al. Two or more rituximab infusions (up to 5) were administered to children who met these criteria.

They demonstrated that remission lasted for six months in half of these children, with better results in older children, longer remission periods

after multiple treatments, and higher response rates for remission without the use of a any oral drug.[52]

In their study, Gulati A et al. demonstrated that rituximab therapy was safe and successful in causing and sustaining remission in a sizable fraction of patients with challenging SRNS and SDNS.[53]

In a Polish multicenter research, Zachwieja et al. used a single rituximab (375 mg/m2) infusion to treat 30 SRNS children who failed to respond to steroids or CNIs.

They noted that rituximab showed an improvement in the response rate when compared to CNI only treatment (93 vs. 69%, P = 0.09), and resulted in an increase in the ability to wean children off both CNIs (77 vs. 50%, P = 0.01) and steroids (60 vs. 27%, P = 0.005).[54]

In 2021, Abeyagunawardena S et al conducted a retrospective analysis in a tertiary care institution to assess the efficacy of various approaches for the withdrawal of cyclosporine A in 81 patients following a 5-year course of treatment.

They noted that levamisole and MMF treated groups showed the maintenance of a sustained remission rate of 80.6% and 82.6% respectively, after withdrawing CsA at one year in comparison to 51.9% in the group given no treatment. The number of relapses decreased with levamisole by 2.49 times as compared to other group (RR= 2.488, p = 0.020, CI 95%) and treatment with MMF decreased relapses by 2.77 times as compared to no treatment group (RR=2.769, p = 0.022, CI 95%). Thus, this study showed a benefit of giving either LV or MMF in an attempt to withdraw long term CsA therapy in patients with NS.[55]

Further studies are required to test the efficacy of MMF, cyclophosphamide and rituximab in inducing remission and, as part of a combination therapy, achieving sustained remission in patients with SRNS and SDNS.

This study was conducted to observe the various CNI sparing strategies being used in a tertiary care hospital after atleast one year of therapy in SDNS and SRNS patients; who have achieved remission (CR or PR) with the same.

MATERIALS AND METHODS

Study centre: Department Of Nephrology and Paediatrics, DMCH, Ludhiana

Study design: Observational study.

Duration: December 2022 to November 2023, minimum 6 months follow up period for each patient.

Source of data: Source of data was patients of SDNS and SRNS between 3 months and 18 years of age according to the inclusion criteria.

Inclusion criteria:

Patients of steroid resistant (SRNS) and steroid dependent (SDNS) between the age of 3 months to 18 years who had received atleast one year of therapy with either of the calcineurin inhibitors (tacrolimus or cyclosporine) and were in complete or partial remission at the time of initiation of study

Exclusion criteria:

1. Patients with secondary form of nephrotic syndrome, or as part of systemic diseases.
2. Patients who had taken CNI for less than 1 year
3. Patients who had frequent relapses on CNI (≥ 3 relapses)
4. Patients with congenital nephrotic syndrome
5. Patients who refused for giving consent for the study

Consent

Written consent was taken from the parents of the children. Patients were assessed in detail and data was collected on specified patient proforma including information regarding demographic profile, type of nephrotic syndrome, drug history, relevant investigations, treatment response, complications (if any) and side effects.

Definitions used:

1. Nephrotic syndrome was defined as: nephrotic range proteinuria; ≥3+ by dipstick, proteinuria ≥ 40 mg/m^2/hr (>1000

mg/m^2/day), urinary protein to creatinine ratio (Up/Uc) ≥2mg/mg; hypoalbuminemia (<3 g/dl); and edema.

2. Steroid resistant nephrotic syndrome : Those who do not achieve remission after 4-6 weeks of therapy with prednisone
3. Steroid-dependent nephrotic syndrome: Patients who suffer from two consecutive relapses on steroid therapy or within 14 days of stopping steroids
4. Frequent relapse was defined as: ≥ 2 relapses in first 6 months from disease onset; ≥ 4 relapses in the 12 months period
5. Relapse: urine protein ≥3+ (urine protein/ urine creatinine ratio >2mg/mg) for 3 consecutive early morning specimens, while being in remission previously

Treatment protocol

Patients of SRNS and SDNS who had already received atleast one year of therapy with any of the CNI (cyclosporine or tacrolimus) were enrolled. Baseline demographic values, laboratory values including eGFR, side effect profile were recorded.

The CNI sparing treatment strategy used by the treating physicians (on their discretion) was recorded for each patient. Data regarding the incidence of sustained remission (steroid free for 6 months), number of relapses, any infection, side effects , corticosteroid and CNI use, dose of each drug, haematological and biochemical test results like serum creatinine, albumin, urine protein creatinine ratio and number of additional drugs used were recorded on regular follow up visits.

Relapse was identified using urine dipstick and confirmed by the measurement of urine protein to creatinine ratio on a spot urine sample.

Relapses were managed with prednisolone at 2 mg/kg/day until remission, followed by 1.5 mg/kg on alternate days for 4 weeks.

Treatment failure was defined as:

1. Recurrence of steroid resistance (no response after relapse, after 4 weeks full dose steroids)

2. Frequent relapses (≥ 2 relapses in six months/ ≥ 3 in 12 months)
3. Irreversible increase in serum creatinine (eGFR < 50 ml/min/ 1.73m2)
4. Shifting of patients with steroid dependent to steroid resistant state
5. Failure to taper off calcineurin inhibitor

Method of collection of data

It was an observational hospital based study of patients from Dec 22 to Nov 23 of SDNS and SRNS who were already on any one of the calcineurin inhibitors, were either in partial or complete remission and were started on any of the CNI sparing drug. Patients were followed up for a minimum period of 6 months and upto maximum 12 months.

Outcome

The primary objective was the incidence of sustained remission after change in therapy

The secondary objectives were

a) the incidence of relapses after the change in therapy
b) the side effect profile after change in therapy

STATISTICAL ANALYSIS

Data were described in the terms of range; mean ± standard deviation (± SD), Median (IQR), frequencies (number of cases) and relative frequencies (percentages) as and where appropriate. To determine whether the data were normally distributed, a Kolmogorov-Smirnov test was used. Comparison of quantitative variables between the study groups was done using Paired t-test. A probability value (*p* value) less than 0.05 was considered statistically significant. All statistical calculations were done using (Statistical Package for the Social Science) SPSS 21.0 version (SPSS Inc., Chicago, IL, USA) statistical program for Microsoft Windows.

OBSERVATIONS AND RESULTS

The study was conducted retrospectively and prospectively to include SRNS and SDNS patients attending outpatient clinic in Nephrology and Paediatrics Department. A total number of 29 patients were enrolled as per the inclusion criteria and followed up for 6-12 months.

1. **Distribution according to the type of nephrotic syndrome: steroid dependent or steroid resistant nephrotic syndrome (SDNS/SRNS)**

Table 1: Distribution of study participants according to the diagnosis: (SDNS/SRNS)

Diagnosis (FRNS/SDNS/SRNS)	No. of cases	Percentage
SDNS	18	62.1%
SRNS	11	37.9%
Total	29	100.0%

Out of the total 29 patients, 18 patients were having steroid dependent nephrotic syndrome and 11 were having steroid resistant nephrotic syndrome.

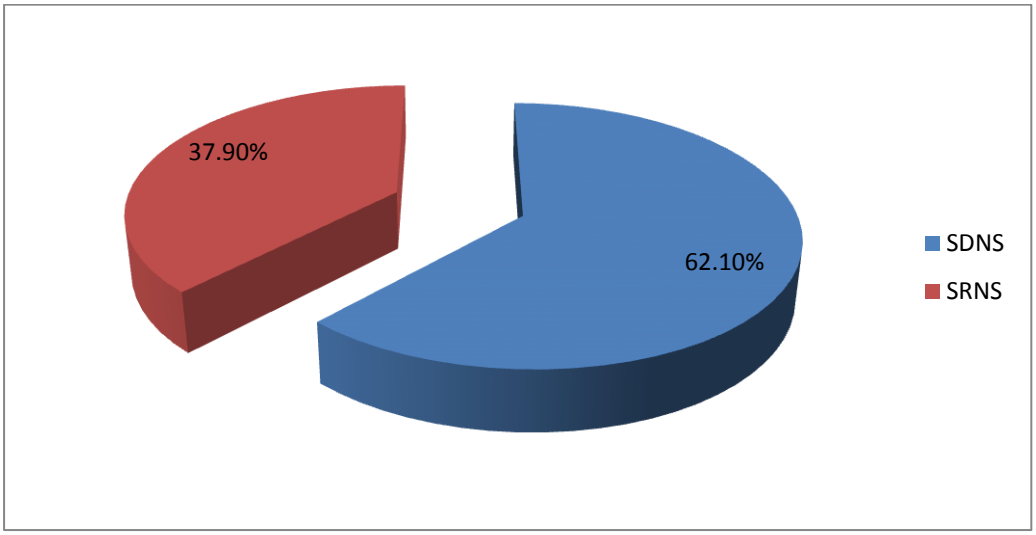

Figure 1: Distribution of study participants according to the diagnosis

2. Demographic Data

A) Regional background of the study population

Table 2: Regional distribution of study participants

Urban/Rural	No. of cases	Percentage
URBAN	16	55.2%
RURAL	13	44.8%
Total	29	100.0%

In the study group, 16 patients were from urban background and 13 were from rural background

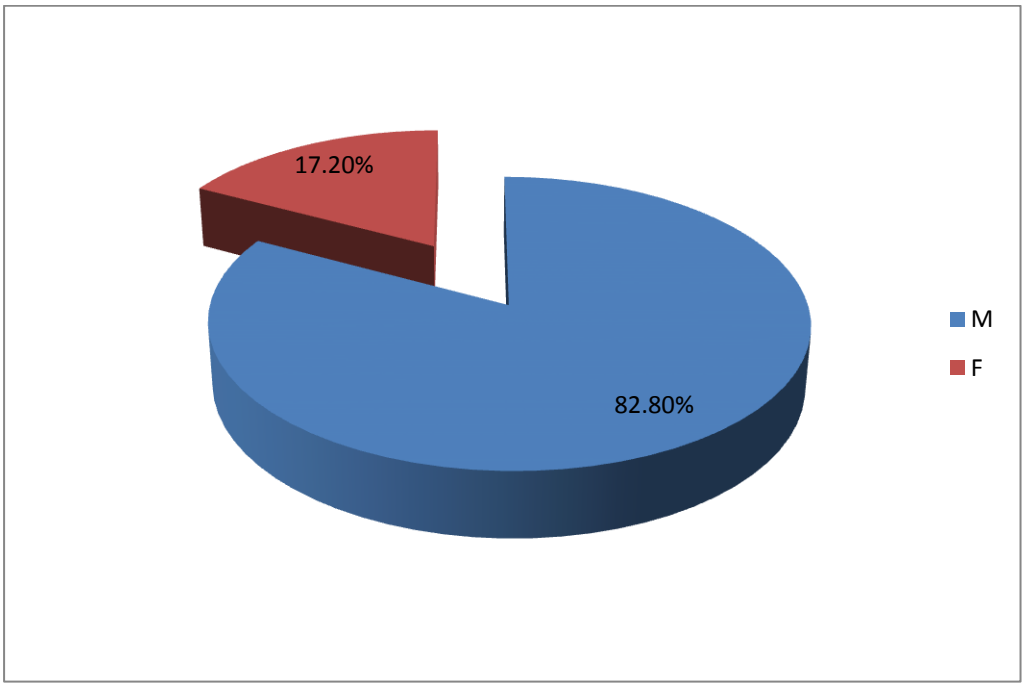

Figure 2: Distribution of study participants based on regional background

B) Gender

Table 3: Distribution of study participants based on Gender

SEX (M/F)	Total no. of cases	Percentage	SDNS	Percentage	SRNS	Percentage
M	24	82.8%	16	88.9%	8	72.7%
F	5	17.2%	2	11.1%	3	27.3%
Total	29	100.0%	18	100%	11	100%

Among the study participants, 24 (i.e. 82.8%) children were male and 5 were female; showing male predominance in our study.

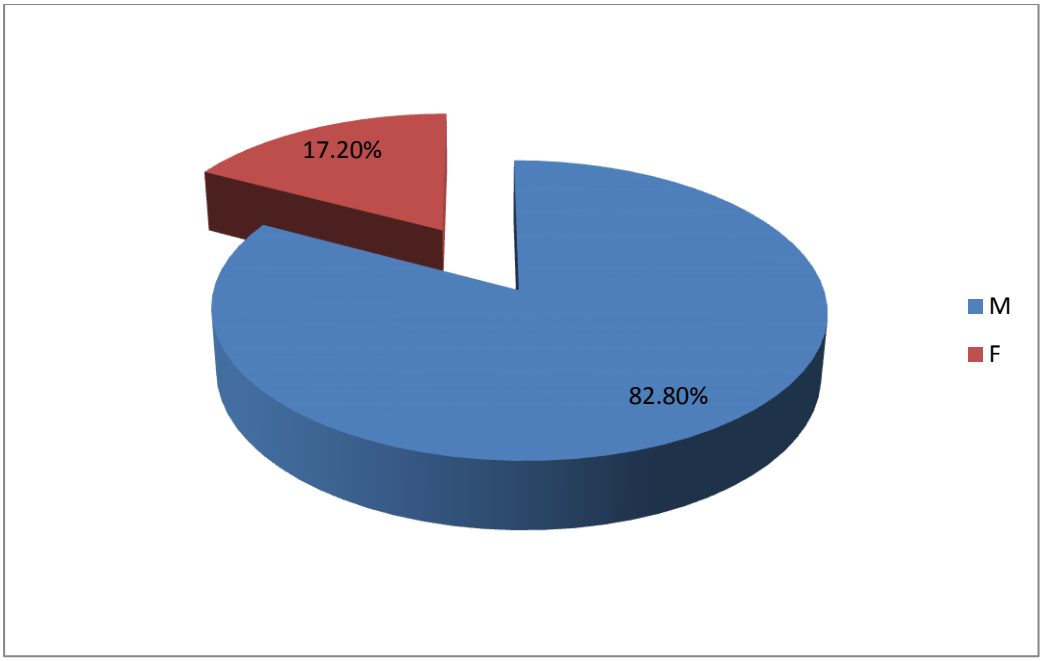

Figure 3: Distribution of study participants based on Gender

C) Age of onset of nephrotic syndrome

Table 4: Distribution of patients according to the age of onset of nephrotic syndrome

Age Of Onset (years)	No. of cases	Percentage	SDNS	Percentage	SRNS	Percentage
<1	2	6.9%	2	11.1%	0	0
1-5	26	89.7%	15	83.3%	11	100%
>5	1	3.4%	1	5.6%	0	0
Total	29	100.0%				

The majority of children in the study group had disease onset between 1-5 years of age. 83.3% of SDNS and 100% children with SRNS had disease onset between 1-5 years of age.

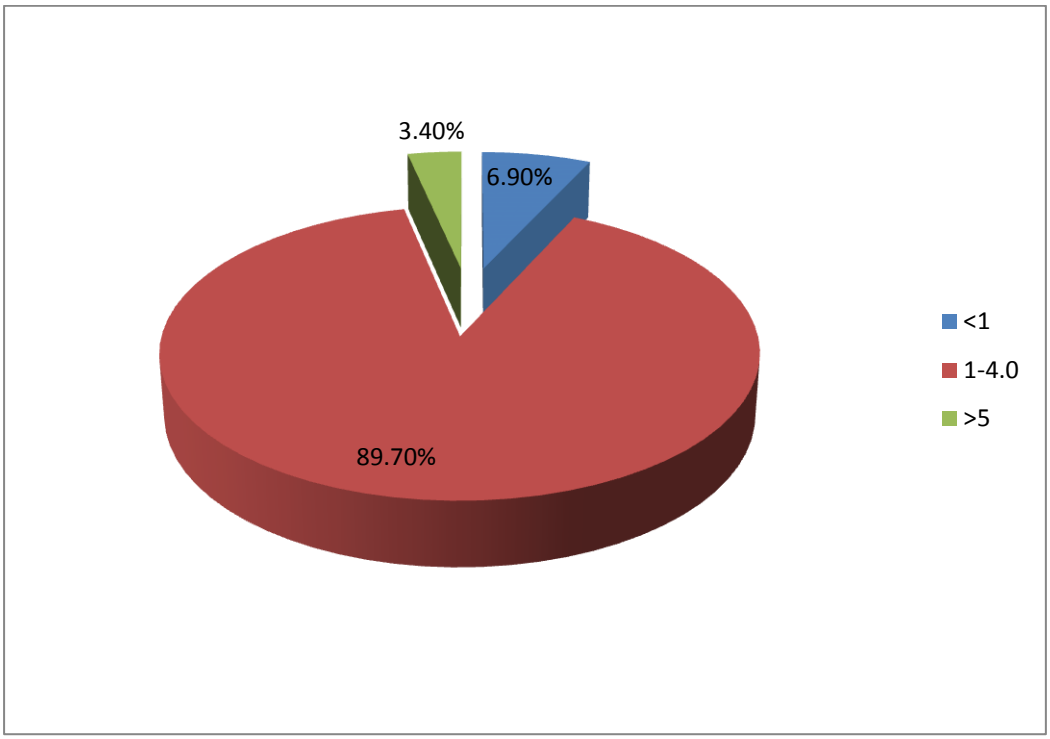

Figure 4: Distribution of patients according to the age of onset of nephrotic syndrome

Table 5: Mean age of onset of NS and interquartile range

	N	Mean	SD	Minimum	Maximum	Median	IQR
Age Of Onset (years)	29	2.77	1.61	0.60	7.50	2.60	1.455-4

The mean age of onset of nephrotic syndrome was 2.77 years and the interquartile range was 1.45- 4 years

3. Associated features :

Table 6: Distribution of study population on the basis of associated features at the time of diagnosis

	No. of cases	Percentage	SDNS	Percentage	SRNS	Percentage
Hypertension	10	34.5%	5	27.8%	5	45.5%
Microscopic haematuria	3	10.3%	1	5.6%	2	18.2%

Out of 29 children, 10 (34.5%) were having hypertension (B.P. readings above 95th percentile for age) at the onset of nephrotic syndrome.

Out of 29 children, 3 (10.3%) were having microscopic haematuria at the onset of nephrotic syndrome.

More children with hypertension and microscopic haematuria at onset turned out to be steroid resistant.

4. Renal biopsy findings

Table 7: Distribution of patients according to renal biopsy findings

Renal biopsy findings if done	No. of cases	Percentage	SDNS	SRNS
NOT DONE	19	65.5%	13	6
MCD	6	20.7%	3	3
FSGS	3	10.3%	1	2
PODOCYTOPATHY (not specified)	1	3.4%	1	0
Total	29	100.0%		

Kidney biopsy was not done in majority, i.e. 65.5% of the patients. Out of 10 patients in whom kidney biopsy was done, 6 were diagnosed as having minimal change disease, 3 had FSGS and 1 had podocytopathy (not specified as electron microscopy was not performed).

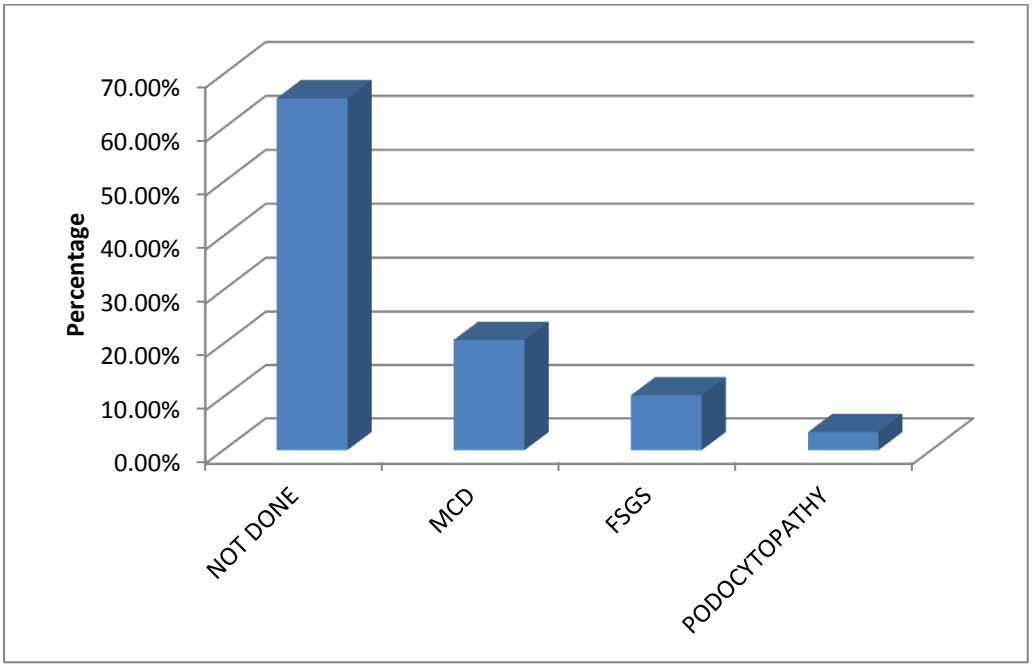

Figure 5: showing the distribution according to kidney biopsy findings

5. **Distribution of patients on the basis of steroid use at the time of enrolment**

Out of 29 patients, 13 were being given steroids at the time of enrolment into our study.. All these patients were steroid dependent. None of the patients in steroid resistant category were on steroids.

6. **Steroid dependent dose :**

Table 8: Steroid dependent dose at the time of enrolment into the study

	N	Mean	SD	Minimum	Maximum	Median	IQR	
Steroid Dependent Dose (mg/kg/day)	13	1.13	0.54	0.20	2.00	1.00	0.75	1.50

The mean steroid dependent dose was 1.13 mg/kg/day at the time of enrolment into the study

7. **Features of steroid toxicity**

Table 9: Distribution of study participants on the basis of presence of features of steroid toxicity

Feature	No. of cases (N=29)	Percentage	SDNS (N=18)	Percentage	SRNS (N=11)	Percentage
Cushingoid features	3	10.3%	2	11.1%	1	9%
Cataract	2	6.9%	2	11.1%	0	
Short stature	1	3.4%	1	5.5%	0	
Obesity	4	13.7%	3	16.6%	1	9%

Features of steroid toxicity were seen in 10 patients at the time of enrolment into our study, the majority of these patients were in the SDNS category.

8. Immunosuppression before CNI

Table 10: Distribution of cases as per the number of immunosuppressive agents used in the past before giving CNI:

No. of Prior Immunosuppressive	No. of cases	Percentage
0	14	48.2%
1	9	31.0%
2	2	6.8%
3	4	13.7%
Total	29	100.0%

It was seen that in the majority i.e. 48.2 % patients, calcineurin inhibitor was the first immunosuppressive used; while in others, immunosuppressive agents like levamisole, cyclophosphamide, mycophenoalte mofetil had been given earlier.

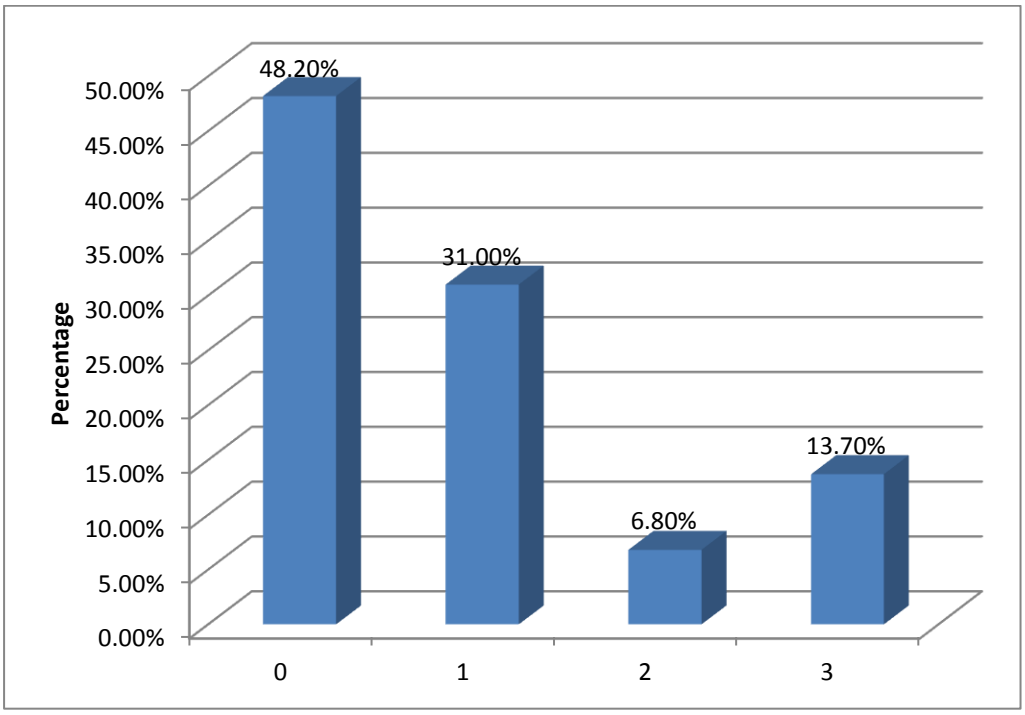

Figure 6: Distribution of cases according to the number of immunosuppressive agents used before giving CNI

Table 11: Nature of previous immunosuppressive agent used:

Drug	SDNS (N=18)	Percentage	SRNS (N=11)	Percentage
Levamisole	6	33.3%	1	9.1%
Cyclophosphamide	4	22.2%	5	45.5%
Mycophenolate mofetil	8	44.4%	1	9.1%

Mycophenolate and cyclophosphamide were the most frequenty used immunosuppressive agents used prior to the use of CNI; MMF was used more in SDNS patients while cyclophosphamide was used most frequently in SRNS patients.

9. Calcineurin inhibitor being used :

Table 12: Distribution of cases according to calcineurin inhibitor being given prior to change in therapy:

CNI being given	Total no. of cases	Percentage	SDNS	Percentage	SRNS	Percentage
TACROLIMUS	25	86.2%	16	88.8%	9	81.8%
CYCLOSPORINE	4	13.8%	2	11.1%	2	18.1%
Total	29	100%	18	100.0%	11	100%

It was seen that majority i.e. 86.2% of patients were being managed with tacrolimus and only 13.8 % were on cyclosporine. 88.8% of SDNS and 81.8% of SRNS patients were on tacrolimus.

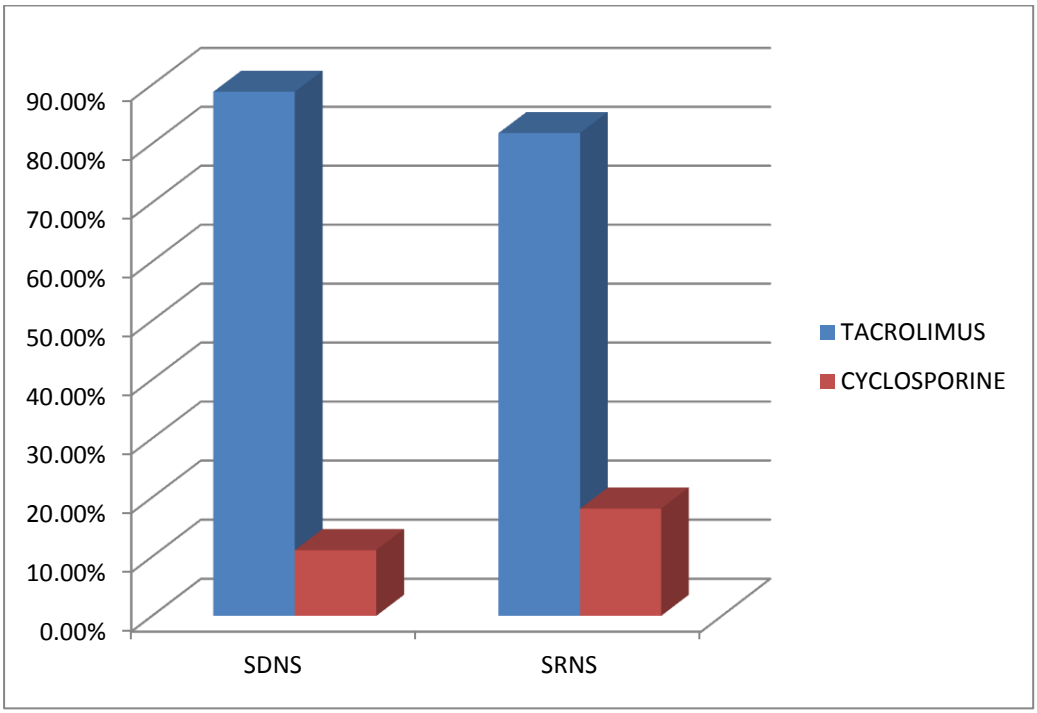

Figure 7: Distribution of SDNS and SRNS patients according to the CNI being given

10. **Duration of calcineurin inhibitor received till the time of enrolment into the study**

Table 13: Duration of CNI therapy already given to the patients

	N	Mean	SD	Minimum	Maximum	Median	IQR
DURATION OF CNI THERAPY (years)	29	2.95	0.86	1.58	4.75	3.00	2.08-3.54

The patients enrolled in our study were on CNI therapy for atleast 12 months and for a mean period of 2.9 years.

11. **Dose of calcineurin inhibitor being used at the beginning of study:**

As already seen, more patients were on tacrolimus than cyclosporine. The mean dose was 0.12 (SD 0.06) mg/kg/day for tacrolimus and 3.72 (SD 2.64) mg/kg/day for cyclosporine.

12. **Incidence relapse rate on CNI therapy (over the last 12 months prior to enrolment into our study)**

Table 14: Distribution of participants on the basis of number of relapses on CNI therapy

	N	Mean	SD	Minimum	Maximum	Median	IQR
No of Relapses in last One Year (on CNI)	29	1.55	0.99	0.00	4.00	2.00	1-2

The incidence relapse rate on CNI therapy was 1.55 with an interquartile range of 1-2.

13. Side effects of CNI therapy prior to enrolment into our study:

Table 15: Incidence of side effects to CNI therapy in the study participants

Side effect	No. of cases (N=29)	Percentage
New onset hypertension	6	20.7%
Diarrhoea (with tacrolimus)	2	6.9%
Skin infections	2	6.9%
Transient transaminitis	1	3.4%
UTI	1	3.4%
Acute kidney injury	1	3.4%
Major infection requiring hospitalisation	1	3.4%
Hyperkalemia	0	0
Seizure	0	0
Increased blood sugars	0	0
Hypertrichosis	0	0
Gingival hyperplasia (with cyclosporine)	0	0

It was observed that new onset hypertension was the most frequent side effect observed with CNI; seen in 6 (20.7%) patients, followed by diarrhoea, skin infections seen in 2 (6.9%) patients each.

14. Status of disease activity/remission

Table 16: Distribution of patients according to the status of nephrotic syndrome at the start of study; i.e. complete/partial remission

Status of patients at the start of study (CR/PR)	Total no. of cases (N=29)	Percentage	SDNS (N=18)	Percentage	SRNS (N=11)	Percentage
Complete remission	22	75.9%	16	88.9%	6	54.5%
Partial remission	7	24.1%	2	11.1%	5	45.5%
Total	29	100.0%	18		11	

It was observed that in the study group, 22 patients were in complete remission and 7 were in partial remission at the time of enrolment. The majority, i.e.16 (88.9%) patients with SDNS while only 6 (54.5%) patients with SRNS were in complete remission at the time of enrolment into the study. The remaining patients were in partial remission at the time of enrolment.

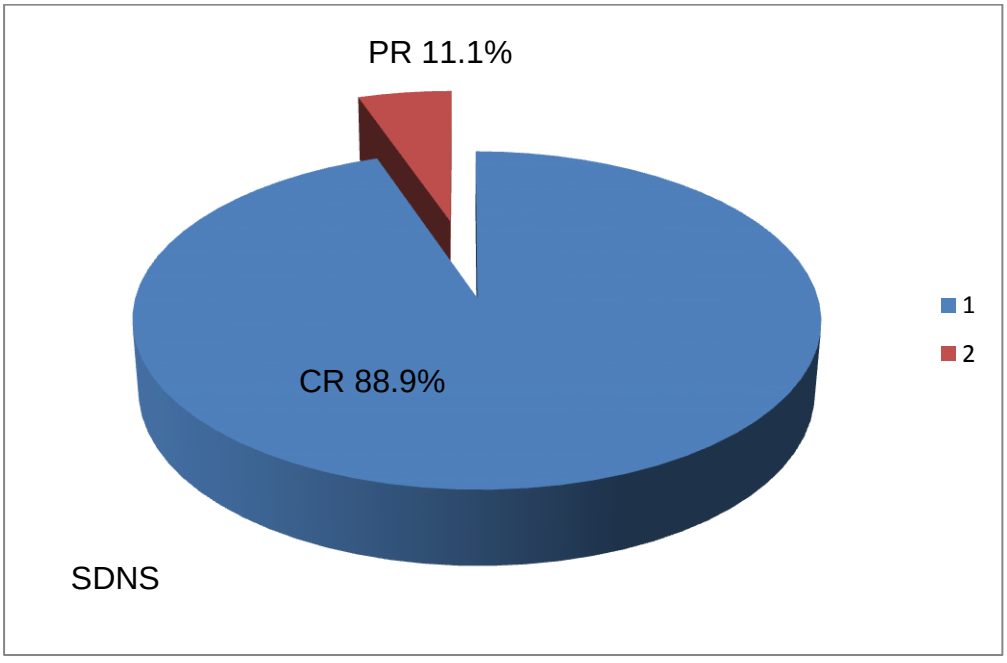

Figure A: SDNS

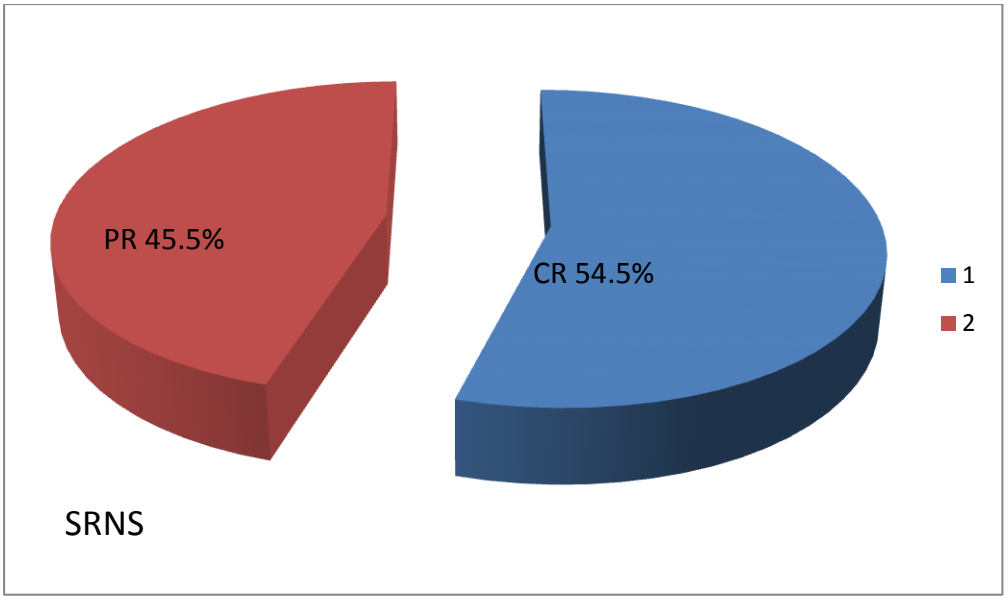

Figure B: SRNS

Figure 8: Status of remission of nephrotic syndrome at start of study in SDNS (Figure A) and SRNS (Figure B) patients respectively

15. **Age at the time of enrolment in our study i.e. at the time of starting CNI sparing agent**

Table 17: The mean age at the start of CNI sparing agent (i.e. mean age at the start of study):

	N	Mean	SD	Minimum	Maximum	Median	IQR
Age at start of study (years)	29	7.29	2.30	4.25	12.00	7.00	5.59-9.33

The mean age of the patients at the beginning of study i.e. when CNI sparing agent was started, was 7.29 years with an IQR 5.5-9.3 years.

16. Baseline characteristics and investigations of the study participants:

Table 18: Baseline characteristics and investigations of the study participants

	N	Mean	SD	Minimum	Maximum	Median	IQR
Weight (kgs)	29	25.06	7.37	16.00	40.00	22.70	19.6-30.35
Height (cms)	29	116.42	12.74	97.50	144.00	114.00	104.5-126
BMI(kg/m2) in patients more than 5 years age	28	18.40	2.72	15.06	25.00	17.45	16.93-19.3825
Hb(gm/dl)	29	12.03	1.03	10.60	15.00	12.00	11.2-12.4
TLC (/mm3)	29	8559.31	2292.23	5500.00	14400.00	8300.00	6650-10210
Platelet count/mm3	29	305931.03	116234.66	185000.00	651000.00	267000.00	212500-350000
Urine PCR	29	0.53	1.14	0.10	6.15	0.20	0.125-0.35
S. albumin (g/dl)	29	3.80	0.45	3.00	4.80	3.80	3.55-4.11
S.Creat(mg/dl)	29	0.46	0.23	0.20	1.20	0.40	0.3-0.6
SGOT	29	21.41	7.36	12.00	44.00	19.00	16-24
SGPT	29	19.00	5.86	10.00	32.00	18.00	14-22

17. Strategy employed:

Table 19: Distribution of study participants as per the different strategies employed:

Strategy employed	No. of cases	Percentage
RITUXIMAB	19	65.5%
ORAL CYCLOPHOSPHAMIDE	6	20.7%
METHOTREXATE	2	6.9%
MYCOPHENOLATE	1	3.4%
DECREASED DOSE OF TACROLIMUS	1	3.4%
Total	29	100.0%

It was observed that the majority of patients i.e. 65.5% were given Rituximab as a CNI sparing agent f/b oral cyclophosphamide in 6 (20.7%), methotrexate in 2, MMF in 1 and just the dose of tacrolimus was reduced in 1 patient.

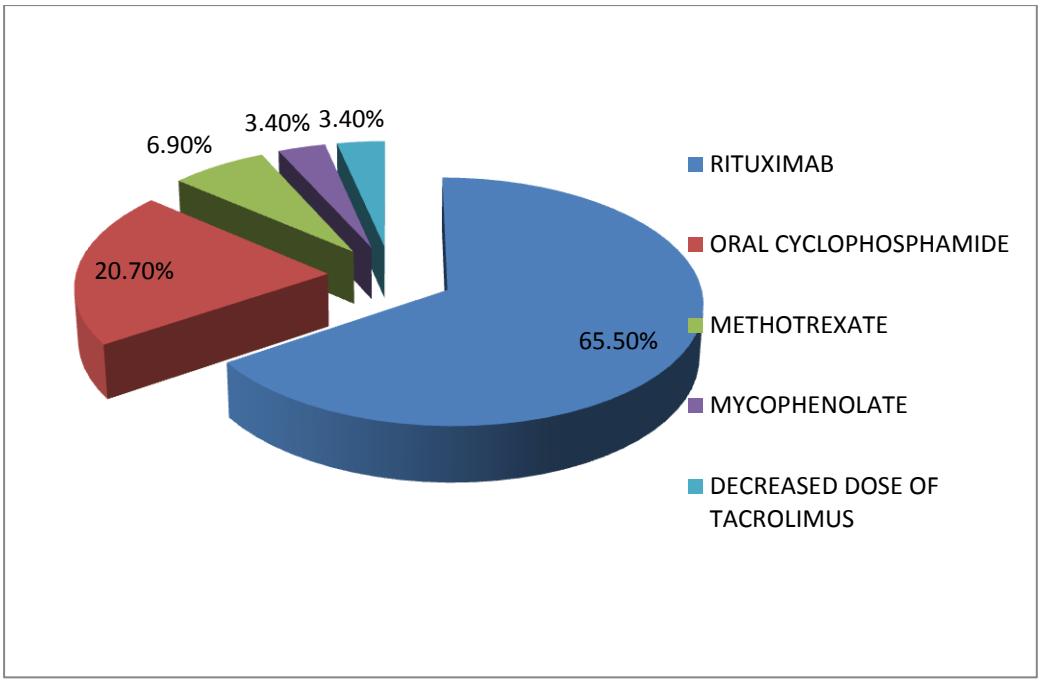

Figure 9: Distribution of study participants as per the different strategies employed

Table 20: Distribution according to strategy employed in SDNS and SRNS patients

Strategy employed	SDNS (N=18)	Percentage	SRNS	Percentage
Rituximab	9	50%	10	90.9%
Oral cyclophosphamide	5	27.7%	1	9 %
Methotrexate	2	11.1%		
Mycophenolate mofetil	1	5.5%		
CNI Dose reduction	1	5.5%		

Rituximab was given to 50% of SDNS and 90.9% of SRNS patients as a CNI sparing therapy.

Whereas oral cyclophosphamide was given to 5 and 1 patient respectively in SDNS and SRNS category. Other agents used in SDNS were methotrexate, mycophenolate mofetil to one patient each.

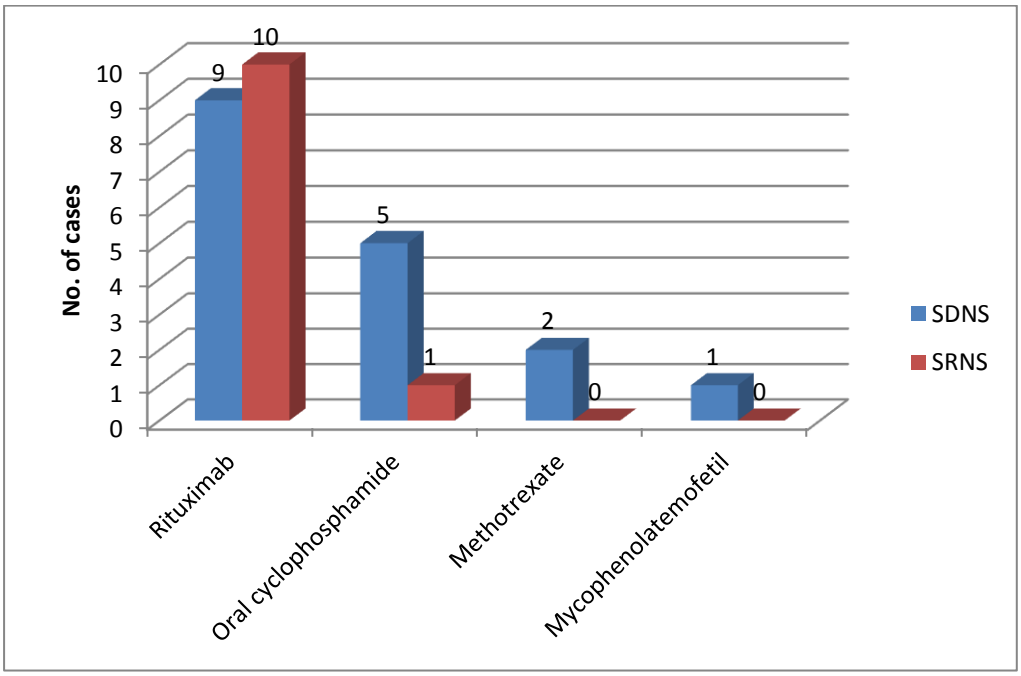

Figure 10: Depicting the number of cases of SDNS and SRNS managed with different drugs

18. Dose of the new drug given:

a) Rituximab:

As per the study protocol, Rituximab was given as infusions of 375 mg/m^2 each

Number of rituximab doses: 13 patients (i.e. 44.8%) were given 3 infusions, 5 were given 2 infusions and one patient was given 4 doses of rituximab. After a single week course of 2 doses of Rituximab, one or two doses of Rituximb were repeated after 6 months in some patients who showed relapse/ increase in proteinuria. This decision was based on the discretion of the treating physician.

Table 21: Distribution according to the number of doses of Rituximab given

2 DOSES	5	17.2%
3 DOSES	13	44.8%
4 DOSES	1	3.4%

b) Cyclophosphamide: It was given orally at a dose of 2 mg/kg/day for a maximum of 12 weeks (cumulative dose not exceeded by 168 mg/kg).

c) Methotrexate: It was given as 10 mg weekly dose

d) Mycophenolate mofetil was given to one patient of SDNS at a dose of 1200 mg/m^2, while gradually decreasing the dose of tacrolimus.

19. Distribution of patients according to the use of additional medications:

Table 22: Distribution of patients according to the use of additional medications

Associated medications	No. of cases (N=29)	Percentage	SDNS (N=18)	Percentage	SRNS (N=11)	Percentage
ACE INHIBITOR	15	51.7%	5	27.7%	10	90.9%
THYROXINE	2	6.89%	0	0	2	18.1%
AMLODIPINE	1	3.4%	0	0	1	9%

In addition to steroids and calcineurin inhibitor, ACE inhibitors were being given to 15 patients (12 – enalapril, 3 - ramipril). The majority of patients on ACE inhibitors were steroid resistant.

1 patient (SRNS) was on 2 antihypertensive agents, enalapril and amlodipine at the time of enrolment in the study. Thyroxine was being given to two patients (6.89%), both SRNS.

20. Follow up

Patients were followed up for a minimum of 6 months and maximum of 12 months period. In the SDNS category, 16 patients were followed up for 6 months and 2 for 12 months. In SRNS category, 9 patients were followed up for a duration of 6 months and 2 were followed for 12 months period.

21. Response to therapy in terms of remission post change in therapy:

Table 23: Remission status at six months post change in therapy:

		Diagnosis (SDNS/SRNS)				Total
		SDNS		SRNS		
Status of patients at 6 months post change in therapy(CR/PR/NR)	CR	14	77.8%	8	72.7%	22
	PR	3	16.7%	3	27.3%	6
	NR	1	5.6%	0	0%	1
Total		18	100.0%	11	100.0%	29

At the end of 6 months follow up, patients with SDNS showed complete remission in 14 (77.8%), partial remission in 3 1(16.7%) and no remission in 1 (5.6%) patient.

In the SRNS category, at the end of 6 months follow up, 8 (72.7%) patients were in complete remission and 3 (27.3%) were in partial remission

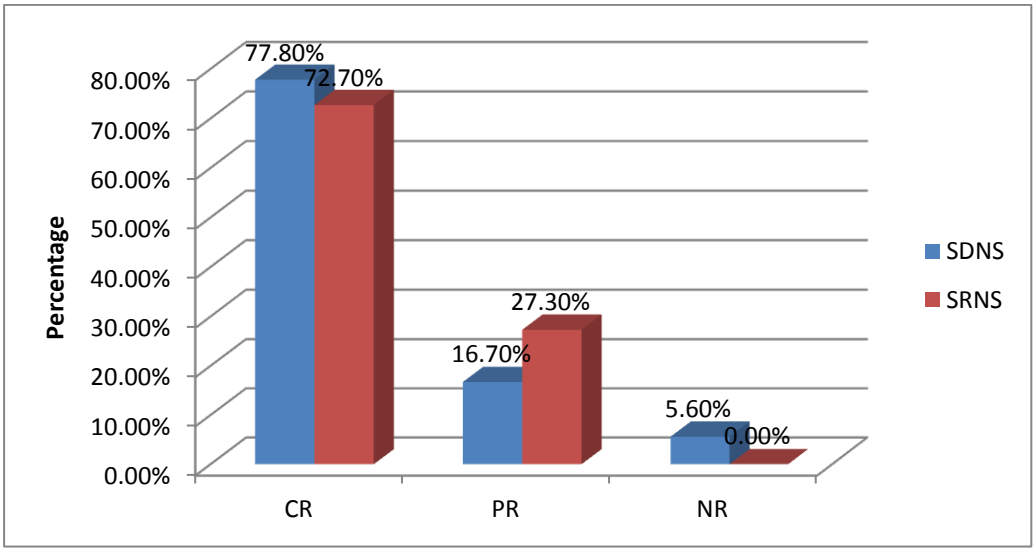

Figure 11: Remission status at six months post change in therapy

Table 24: Remission status at twelve months post change in therapy:

		Diagnosis (SDNS/SRNS)				Total
		SDNS		SRNS		
Status of patients at 12 months post change in therapy(CR/PR/NR)	CR	14	87.5%	8	88.8%	22
	PR	1	6.25%	1	11.1%	2
	NR	1	6.25%	0	0%	1
		16	100.0%	9	100.0%	25

16 patients with SDNS and 9 patients with SRNS were followed for 12 months.

At the end of 12 months, in SDNS category, 14 (87.5%) were in complete remission, 1 (6.25%) was in partial remission while one patient failed to achieve remission even after 12 months follow up.

In the SRNS category, 8 (88.8%) were in complete remission while 1 (11.1%) was in partial remission.

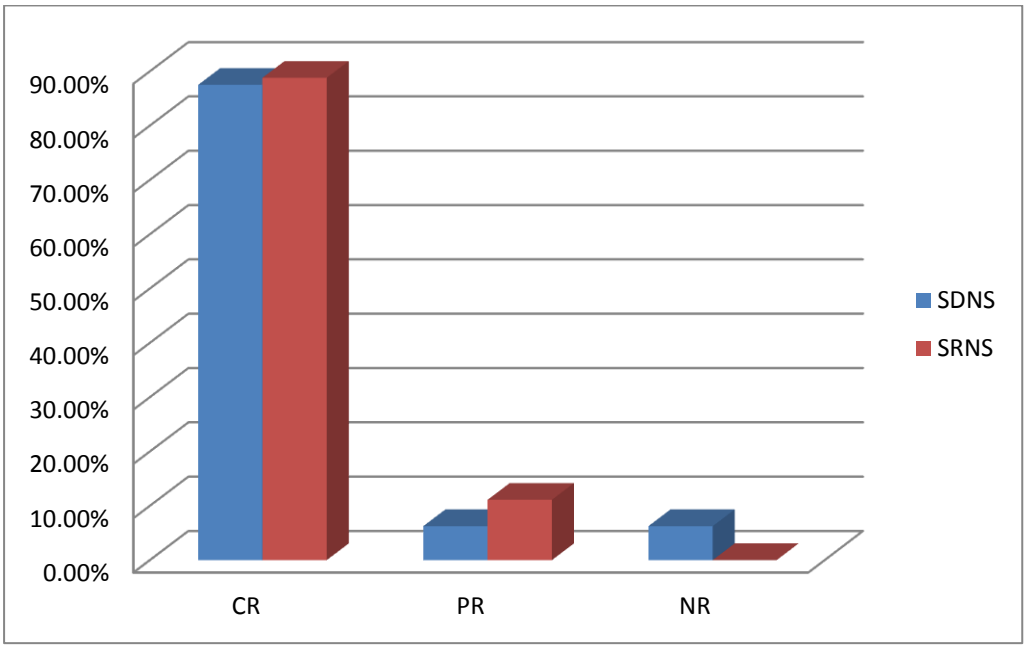

Figure 12: Remission status at twelve months post change in therapy

22. Response of the new strategy employed in terms of steroid free status; and duration of steroid free period

Table 25: steroid free status of the study population

Steroids free (Y/N)	No. of cases	Percentage
Y	28	96.6%
N	1	3.4%
Total	29	100.0%

Table 26: Duration of steroid free period

	N	Mean	SD	Minimum	Maximum	Median	IQR
Duration of steroid free period (in months)	28	6.98	3.96	1.00	12.00	6.00	3-11

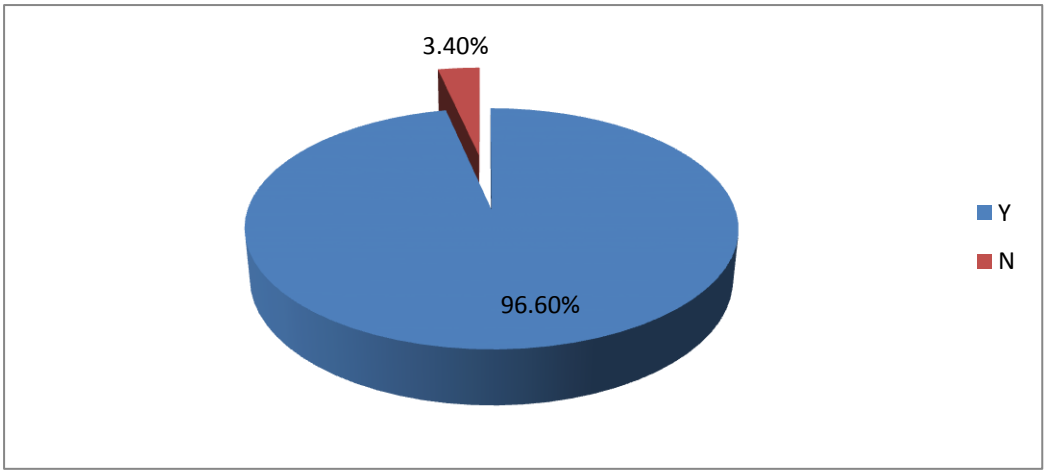

Figure 13: Distribution of patients who remained steroid free

It was observed that 28 (96.6%) patients remained free of steroids for a variable period of time ranging from 1 month to 12 months. The mean duration for which patients remained free of steroids was 6.9 months. Steroid free period was more than six months (i.e. sustained remission) in 16 (i.e. 57.1%) patients.

In one patient with SDNS, in whom steroid could not be withdrawn at all, the dosage was reduced and he was being maintained at a dose of 0.1mg/kg/day

23. Response of the changed therapy in terms of withdrawal of calcineurin inhibitors:

Table 27: Distribution of study participants in terms of CNI withdrawal

CNI free (Y/N)	No. of cases	Percentage
N	17	58.6%
Y	12	41.4%
Total	29	100.0%

It was seen that calcineurin inhibitor was withdrawn in 12 (i.e.41.4%) patients. The mean duration for which CNI were stopped was 4.9 months with an IQR 2-8.75.

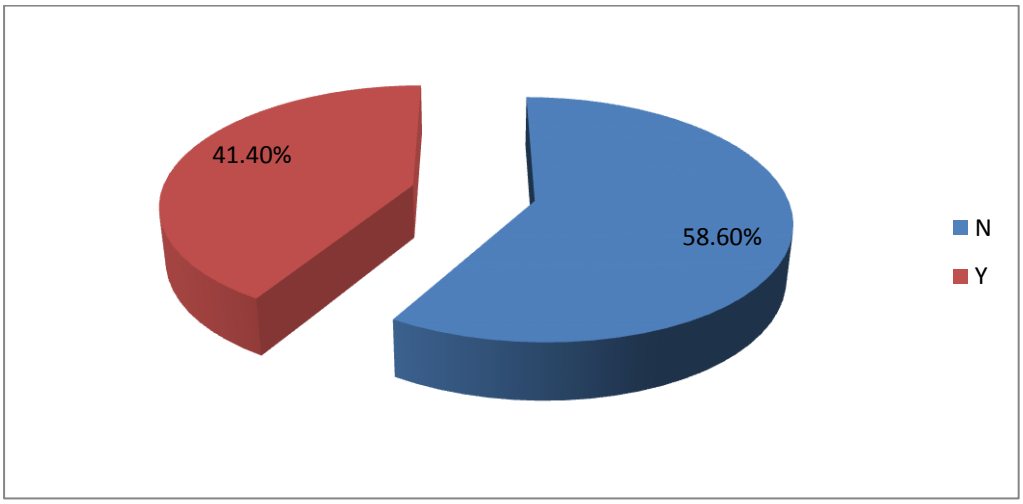

Figure 14: Distribution of study participants in terms of CNI withdrawal

Table 28: CNI free duration after change in therapy

	N	Mean	SD	Minimum	Maximum	Median	IQR
CNI free duration in months)	12	4.92	3.82	1.00	12.00	3.00	2-8.75

Table 29: CNI free duration separately in patients with SDNS and SRNS

		SDNS (N=18)	Percentage	SRNS (N=11)	Percentage	Total
CNI free (Y/N)	Y	9	50.0%	3	27.3%	12
	N	9	50.0%	8	72.7%	17

Table 30: Duration (in months) after which CNI were stopped

CNI stopped after (months)	0	1	5.6%	0	0.0%	1
	3	1	5.6%	1	9.1%	2
	4	1	5.6%	0	0.0%	1
	5	2	11.1%	0	0.0%	2
	6	0		0		
	9	1	5.6%	1	9.1%	2
	10	2	11.1%	0	0.0%	2
	11	1	5.6%	1	9.1%	2
	12	0		0		

In SDNS category, CNI could be withdrawn in 50 % patients, while in SRNS category, CNI could be stopped in only 3 (27.3%) patients.

It was seen that in 5 patients of SDNS, CNI were withdrawn within 6 months of change in therapy, whereas in SRNS, CNI could be withdrawn in only 1 patient in 6 months following the change in therapy.

Table 31: Strategy wise distribution of patients showing the duration (in months) after which CNI were withdrawn

	Strategy employed	N	Mean	SD	Median	IQR		Minimum	Maximum
CNI stopped after (months)	RITUXIMAB	8	8.00	3.16	9.50	5.00	10.75	3.00	11.00
	CYCLOPHOSPHAMIDE	2	2.00	2.83	2.00	0.00	4.00	0.00	4.00
	METHOTREXATE	1	3.00	.	3.00	3.00	3.00	3.00	3.00
	MYCOPHENOLATE	1	9.00	.	9.00	9.00	9.00	9.00	9.00
	Total	12	6.67	3.75	1.08	4.28	9.05	0.00	11.00

CNI were stopped in 2 patients managed with cyclophosphamide (both SDNS) after a mean duration of 2 months.

In 8 patients given rituximab, CNI were stopped after a mean duration of 8 months.

In 1 patient given methotrexate, CNI stopped after 3 months and in another patient, who was given mycophenolate, stopped after 9 months.

Table 32: CNI free period (in months) in different strategies

	Strategy employed	N	Mean	SD	Median	IQR		Minimum	Maximum
CNI free period (in months)	RITUXIMAB	8	3.38	2.97	2.00	1.25	6.00	1.00	9.00
	CYCLOPHOSPHAMIDE	2	10.00	2.83	10.00	8.00	12.00	8.00	12.00
	METHOTREXATE	1	9.00	.	9.00	9.00	9.00	9.00	9.00
	MYCOPHENOLATE	1	3.00	.	.	.	.	3.00	3.00
	Total	12	4.92	3.82	1.10	2.49	7.35	1.00	12.00

The CNI free period was longest with cyclophosphamide; mean duration of 10 months. With the use of Rituximab, it was 3 months.

24. Distribution of patients according to the time (in months) to first relapse after changing therapy from CNI to other agents:

Table 33: Time (in months) to first relapse after change in therapy

	N	Mean	SD	Minimum	Maximum	Median	IQR
First relapse after change in therapy (months)	20	5.60	2.62	1.00	11.00	5.50	4-7.75

It was seen that 9 (31%) out of 29 patients had no relapse in the follow up period.

In the remaining 20 patients, the mean duration to first relapse after change in therapy was 5.6 months.

25. No. of relapses after change in therapy

Table 34: Incidence of relapses post change in therapy in 6 months:

	No. of relapses	In SDNS (N=18)	Percentage	In SRNS (N=11)	Percentage	Total patients
No. of relapses after change in therapy (in 6 months)	0	11	61.1%	6	54.5%	17
	1	6	33.3%	5	45.5%	11
	3	1	5.6%	0	0.0%	1
Total relapses		9		5		

During 6 months follow up period, in patients with SDNS, 11(61.1%) patients ha relapse; 6 (33.3%) patients had 1 relapse and only one patient had 3 relapses.

During 6 months follow up period, among 11 patients with SRNS, 6 (54.5%) pa had no relapse; 5 (45.5%) patients had 1 relapse.

Table 35: Incidence of relapses post change in therapy in 12 months:

	No. of relapses	In SDNS (N=16)	Percentage	In SRNS (N=9)	Percentage	Total
No. of relapses after change in therapy (in 12 months)	0	6	37.5%	3	33.3%	9
	1	6	37.5%	5	55.5%	12
	2	3	18.7%	1	11.1%	4
	3	1	5.5%	0	0.0%	1
Total relapses		15		7		

During 12 months period, 6 (37.5%) patients had no relapse, 6 (37.5%) had 1 relapse, 3 (18.7%) had 2 relapses and 1 (5.5%) had 3 relapses.

During 12 months period, among 9 patients with SRNS, 3 (33.3%) patients had no relapse, 6 (66.6%) had 1 relapse, 1 (18.7%) had 2 relapses.

26. Incidence relapse rate on follow up , post change in therapy :

At 6 months: 0.48 relapses/patient year

At 12 months: 0.88 relapses/patient year

Table 36: Incidence relapse rate before and after change in therapy

	N	Mean	SD	Minimum	Maximum	Median	IQR
No of Relapses in last One Year (on CNI)	29	1.55	0.99	0.00	4.00	2.00	1-2
No. of relapses after change in therapy (in 6 months)	29	0.48	0.69	0.00	3.00	0.00	0-1
No. of relapses after change in therapy (in 12 months)	25	0.88	0.82	0.00	3.00	1.00	0-1

The incidence relapse rate was 1.55 relapses/patient/year on CNI therapy, 0.48 post 6 months of change in therapy and 0.88 post 12 months of change in therapy

Table 37: Number of relapses prior to after change in therapy (in six months)

	N	Mean	SD	Std. Error Mean	P value	Difference			95% Confidence Interval of the Difference	
						Mean	SD	Std. Error Mean	Lower	Upper
No of Relapses in last One Year (on CNI)	29	1.55	0.99	0.18	0.001	1.069	1.163	0.216	0.627	1.511
No. of relapses after change in therapy (in 6 months)	29	0.48	0.69	0.13						

Table 38: Number of relapses prior to after change in therapy (in twelve months)

	N	Mean	SD	Std. Error Mean	p-value	Difference			95% Confidence Interval of the Difference	
						Mean	SD	Std. Error Mean	Lower	Upper
No of Relapses in last One Year (on CNI)	25	1.62	0.98	0.19	0.005	0.731	1.116	0.219	0.280	1.181
No. of relapses after change in therapy (in 12 months)	25	0.88	0.82	0.16						

27. Investigations on follow up of patients:

All the patients were followed up for 6-12 months as per the study protocol. Urine protein creatinine ratio, serum albumin, serum creatinine levels and eGFR were monitored in addition to other tests.

a) Urine protein to creatinine ratio:

Table 39: showing urine PCR values at baseline 3, 6, 9 and 12 months follow up.

	N	Mean	SD	Std. Error Mean	Z	p-value	Difference			95% Confidence Interval of the Difference	
							Mean	Std. Deviation	Std. Error Mean	Lower	Upper
Urine PCR	29	0.53	1.14	0.21	-1.183	0.237	0.12	1.35	0.25	-0.40	0.63
Urine PCR at 3 months	29	0.41	0.67	0.13							
Urine PCR at 6 months	29	0.67	1.46	0.27	-0.071	0.943	-0.14	0.74	0.14	-0.42	0.14
Urine PCR	25	0.31	0.35	0.07	-1.912	0.056	-0.69	1.60	0.32	-1.36	-0.03
Urine PCR at 9 months	25	1.00	1.60	0.32							
Urine PCR at 12 months	25	0.42	0.76	0.15	-1.113	0.266	-0.11	0.87	0.17	-0.47	0.24

It was seen that though mean uPCR value decreased at 12 months of follow up, as compared to baseline value; but the difference was not statistically significant (p value 0.266)

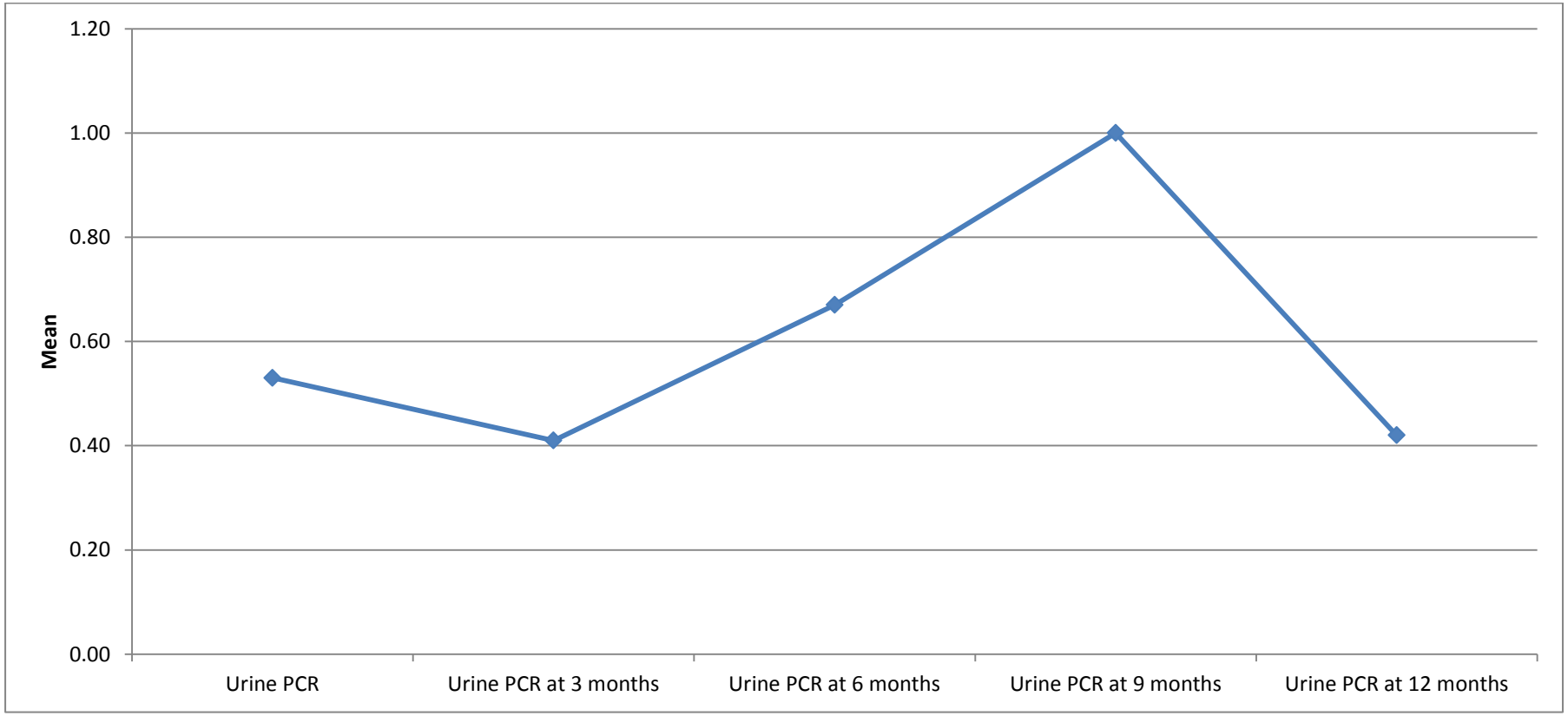

Figure 15: Line diagram showing the uPCR values at baseline and follow up

b) Serum albumin :

Table 40: Depicting the serum albumin levels at baseline and follow up

	N	Mean	SD	Std. Error Mean	Z	p-value	Difference			95% Confidence Interval of the Difference	
							Mean	Std. Deviation	Std. Error Mean	Lower	Upper
S. albumin (g/dl)	29	3.80	0.45	0.08	-0.076	0.939	0.02	0.51	0.10	-0.18	0.21
Serum albumin at 3 months	29	3.79	0.38	0.07							
Serum albumin at 6 months	29	3.77	0.49	0.09	-0.500	0.617	0.03	0.49	0.09	-0.15	0.22
S. albumin (g/dl)	25	3.88	0.42	0.08	-2.212	0.027	0.38	0.81	0.16	0.05	0.71
Serum albumin at 9 months	25	3.50	0.78	0.16							
Serum albumin at 12 months	25	3.87	0.31	0.06	-0.260	0.795	0.01	0.52	0.10	-0.20	0.22

Mean serum albumin increased from 3.80 at baseline to 3.87 gm/dl at 12 months follow up; but again the difference was not significant.

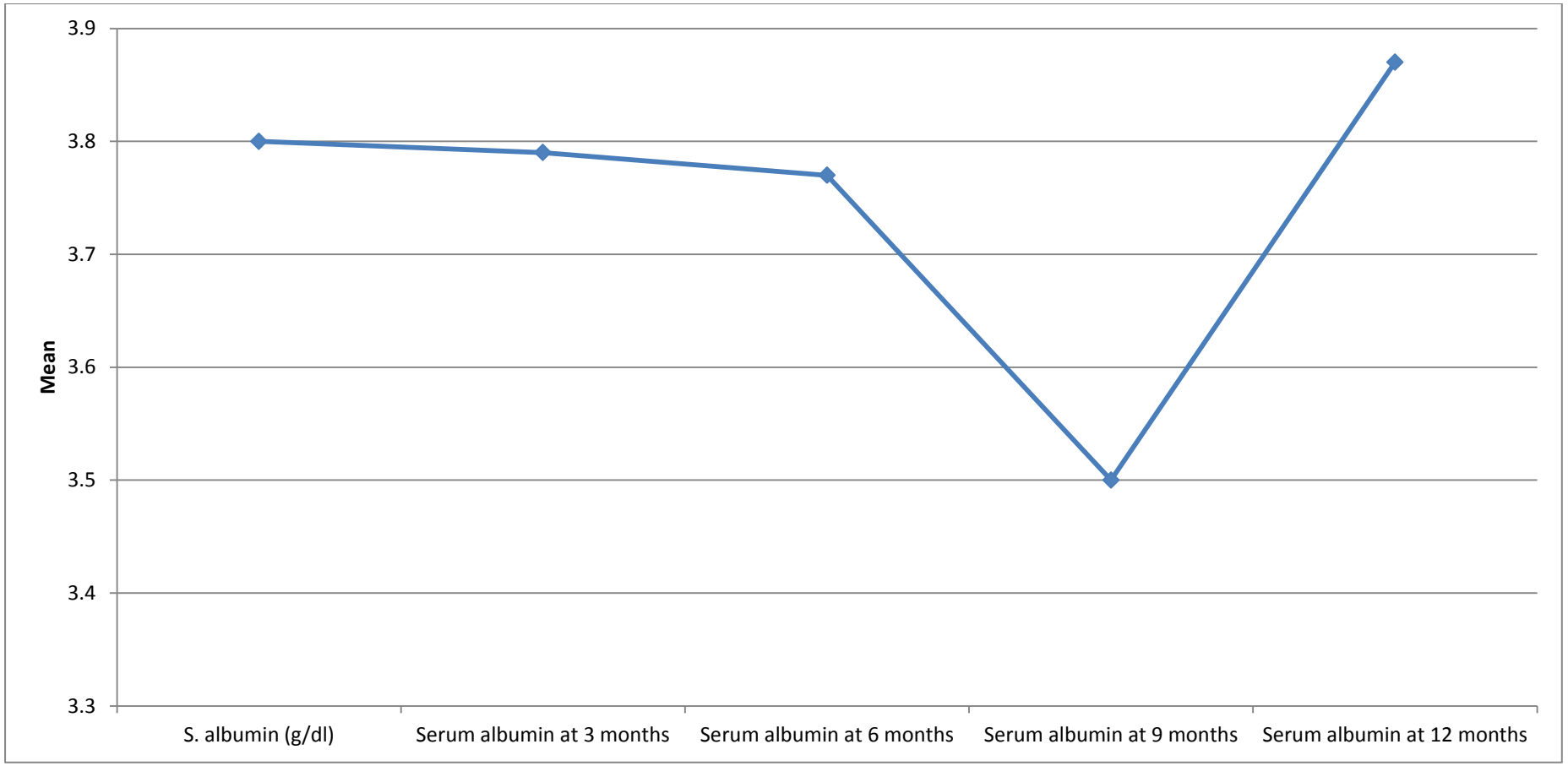

Figure 16: Line diagram depicting the serum albumin levels at baseline and on follow up

c) eGFR

Table 41: Comparison of eGFR before and after change in therapy (in patients with SDNS)

In patients with SDNS, there was an improvement in eGFR at 12 months post change in therapy but it was statistically insignificant.

SDNS	N	Mean	SD	Std. Error Mean	Z	p-value	Difference			95% Confidence Interval of the Difference	
							Mean	Std. Deviation	Std. Error Mean	Lower	Upper
eGFR	18	124.00	57.90	13.65	-1.982	0.064	-19.72	42.22	9.95	-40.72	1.28
eGFR at 6 months	18	143.72	64.23	15.14							
eGFR	16	120.81	59.82	14.95	-2.376	0.031	-43.50	73.22	18.31	-82.52	-4.48
eGFR at 12 months	16	164.31	56.93	14.23							

Table 42: Comparison of eGFR before and after change in therapy (in patients with SRNS)

In patients with SRNS, there was a significant improvement in eGFR at 12 months post change in therapy (p=0.03)

	N	Mean	SD	Std. Error Mean	Z	p-value	Difference			95% Confidence Interval of the Difference	
SRNS							Mean	Std. Deviation	Std. Error Mean	Lower	Upper
eGFR	11	144.27	53.95	16.27	-3.404	0.007	-27.64	26.92	8.12	-45.72	-9.55
eGFR at 6 months	11	171.91	76.37	23.03							
eGFR	9	154.33	51.30	17.10	-1.209	0.261	-25.56	63.44	21.15	-74.32	23.21
eGFR at 12 months	9	179.89	63.95	21.32							

28. Response to different strategies employed:

a) Table 43: Status of patients at 6 months post change in therapy

		Strategy employed										Total
		RITUXIMAB		CYCLOPHOSPHAMIDE		MYCOPHENOLATE		METHOTREXATE		DECREASED DOSE OF TACROLIMUS		
Status of patients at 6 months post change in therapy (CR/PR/NR)	CR	14	73.7%	4	66.7%	1	100.0%	2	100.0%	1	100.0%	22
	PR	4	21.1%	2	33.3%	0	0.0%	0	0.0%	0	0.0%	6
	NR	1	5.3%	0	0.0%	0	0.0%	0	0.0%	0	0.0%	1
Total		19	100.0%	6	100.0%	1	100.0%	2	100.0%	1	100.0%	29

After 6 months post change in therapy, 14 (74.7%) patients (7 SDNS, 7 SRNS) given Rituximab, 4 (66.7%) patients (3 SDNS, 1 SRNS) given cyclophosphamide, 1 patient given MMF (SDNS) both the patients given methotrexate (both SDNS) and 1 patient in whom tacrolimus dose was reduced (SDNS) were in complete remission.

4 patients (21.1%) (1SDNS, 3 SRNS) given Rituximab, 2 patients (SDNS) given oral cyclophosphamide were in partial remission

1 patient (SDNS) given Rituximab did not achieve remission. This patient had FSGS on renal biopsy, achieved remission after third dose of Rituximab at 6 months and stayed in complete remission till 12 months follow up.

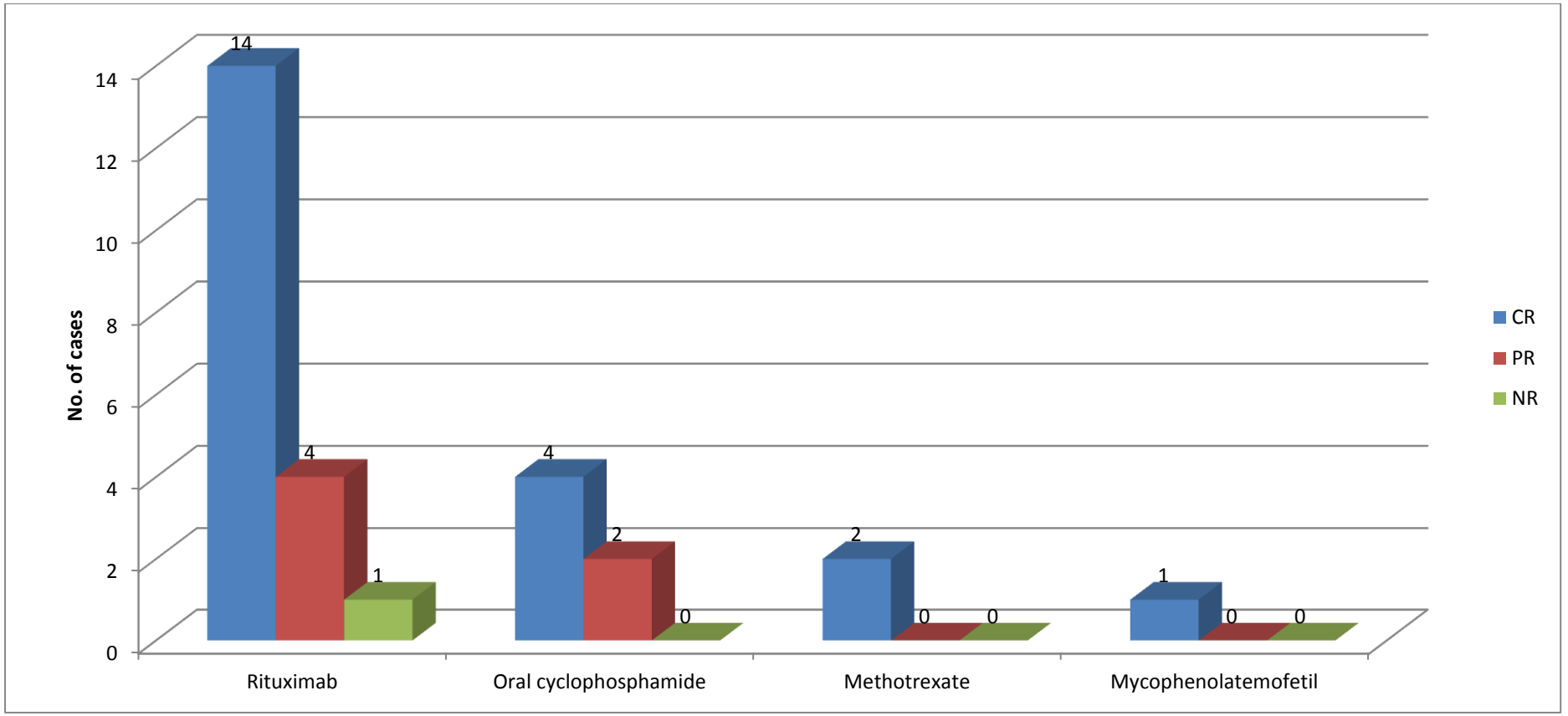

Figure 17: Remission status at 6 months post change in therapy with various drugs

b) Table 44: Status of patients at 12 months post change in therapy

		Strategy implied										Total
		RITUXIMAB		CYCLOPHOSPHAMIDE		MYCOPHENOLATE		METHOTREXATE		DECREASED DOSE OF TACROLIMUS		
Status of patients at 12 months post change in therapy (CR/PR/NR)		2	10.5%	2	33.3%	0	0.0%	0	0.0%	0	0.0%	4
	CR	15	78.9%	3	50.0%	1	100.0%	2	100.0%	1	100.0%	22
	PR	2	10.5%	0	0.0%	0	0.0%	0	0.0%	0	0.0%	2
	NR	0	0.0%	1	16.7%	0	0.0%	0	0.0%	0	0.0%	1
Total		19	100.0%	6	100.0%	1	100.0%	2	100.0%	1	100.0%	29

25 patients out of 29 were followed up for 12 months.

After 12 months post change in therapy, 15 (78.9%) patients (8 SDNS, 7 SRNS) given Rituximab, 3 (50%) patients (2 SDNS, 1 SRNS) given cyclophosphamide, 1 patient given MMF (SDNS), both the patients given methotrexate (both SDNS) and 1 patient in whom tacrolimus dose was reduced (SDNS) were in complete remission.

2 patients (10.5%) (1SDNS, 1 SRNS) given Rituximab were in partial remission

1 patient (SDNS) given oral cyclophosphamide was in non remission at 12 months follow up. This patient had been given earlier levamisole and MMF before CNI. He was on oral tacrolimus for 3 years and had developed features of steroid toxicity (cataract), hypertension on tacrolimus. After oral cyclophosphamide, he had three relapses in one year and was in non remission. CNI could not be stopped in this patient.

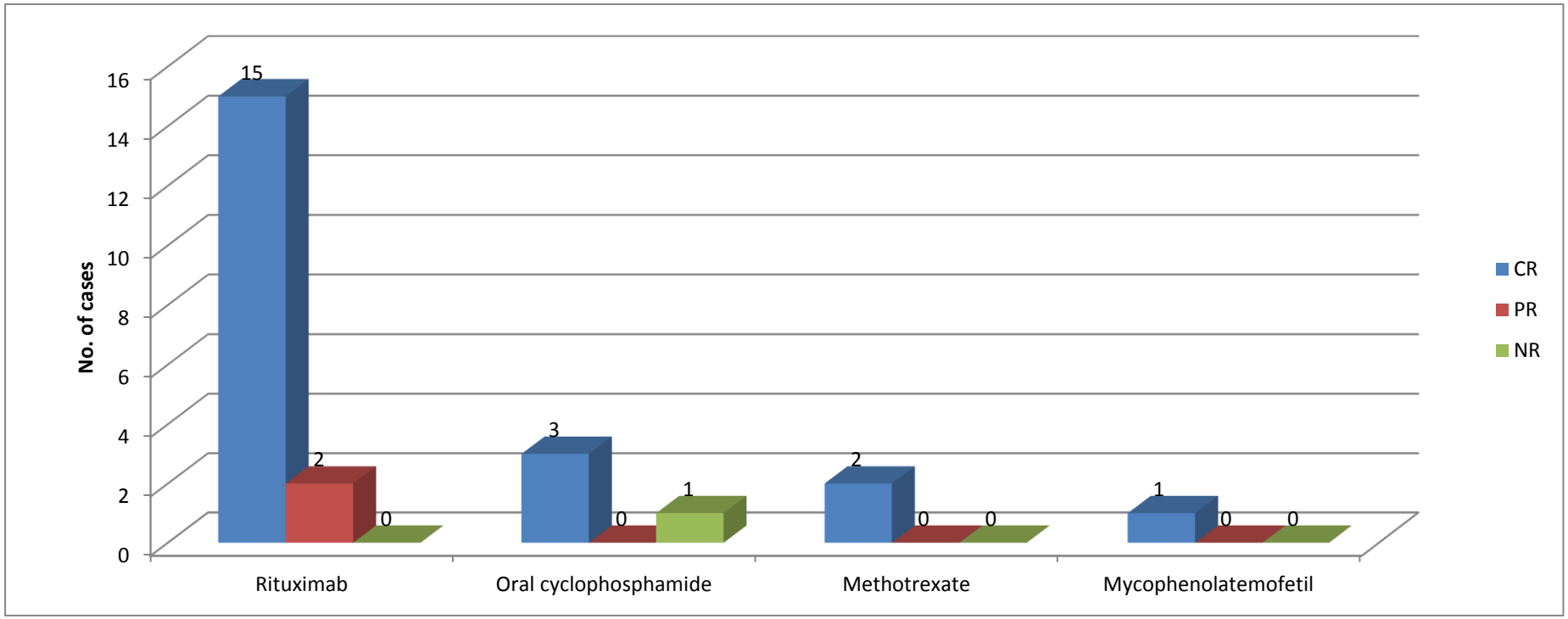

Figure 18: Remission at 12 months therapy with various drugs

c) Table 45: Strategy wise distribution showing steroid free status

		Strategy implied										Total
		RITUXIMAB		CYCLOPHOSPHAMIDE		MYCOPHENOLATE		METHOTREXATE		DECREASED DOSE OF TACROLIMUS		
	Y	19	100.0%	6	100.0%	1	100.0%	1	50.0%	1	100.0%	28
	N	0	0.0%	0	0.0%	0	0.0%	1	50.0%	0	0.0%	1
Total		19	100.0%	6	100.0%	1	100.0%	2	100.0%	1	100.0%	29

All the patients managed with Rituximab, cyclophosphamide, MMF could be made steroid free for a variable period of time (exact mentioned in next table). One patient of SDNS who was given methotrexate remained steroid dependent.

d) Table 46: Duration of steroid free period with each therapy

		N	Mean	SD	median	IQR		Minimum	Maximum
If yes, duration off steroids (in months)	RITUXIMAB	19	7.76	3.88	9.00	4.00	12.00	1.50	12.00
	CYCLOPHOSPHAMIDE	6	3.67	2.50	3.00	1.75	5.75	1.00	8.00
	MYCOPHENOLATE	1	12.00	.	12.00	12.00	12.00	12.00	12.00
	METHOTREXATE	1	4.00	.	4.00	4.00	4.00	4.00	4.00
	DECREASED DOSE OF TACROLIMUS	1	10.00	.	10.00	10.00	10.00	10.00	10.00
	Total	28	6.98	3.96	0.75	5.45	8.52	1.00	12.00

The mean steroid free duration was 7.76 months with Rituximab (n=19), 3.67 months with cyclophosphamide (n=6), 12 months with MMF (but only 1 patient), 4 months with methotrexate (n=2) and 10 months in one SDNS patient in whom tacrolimus dose was being reduced

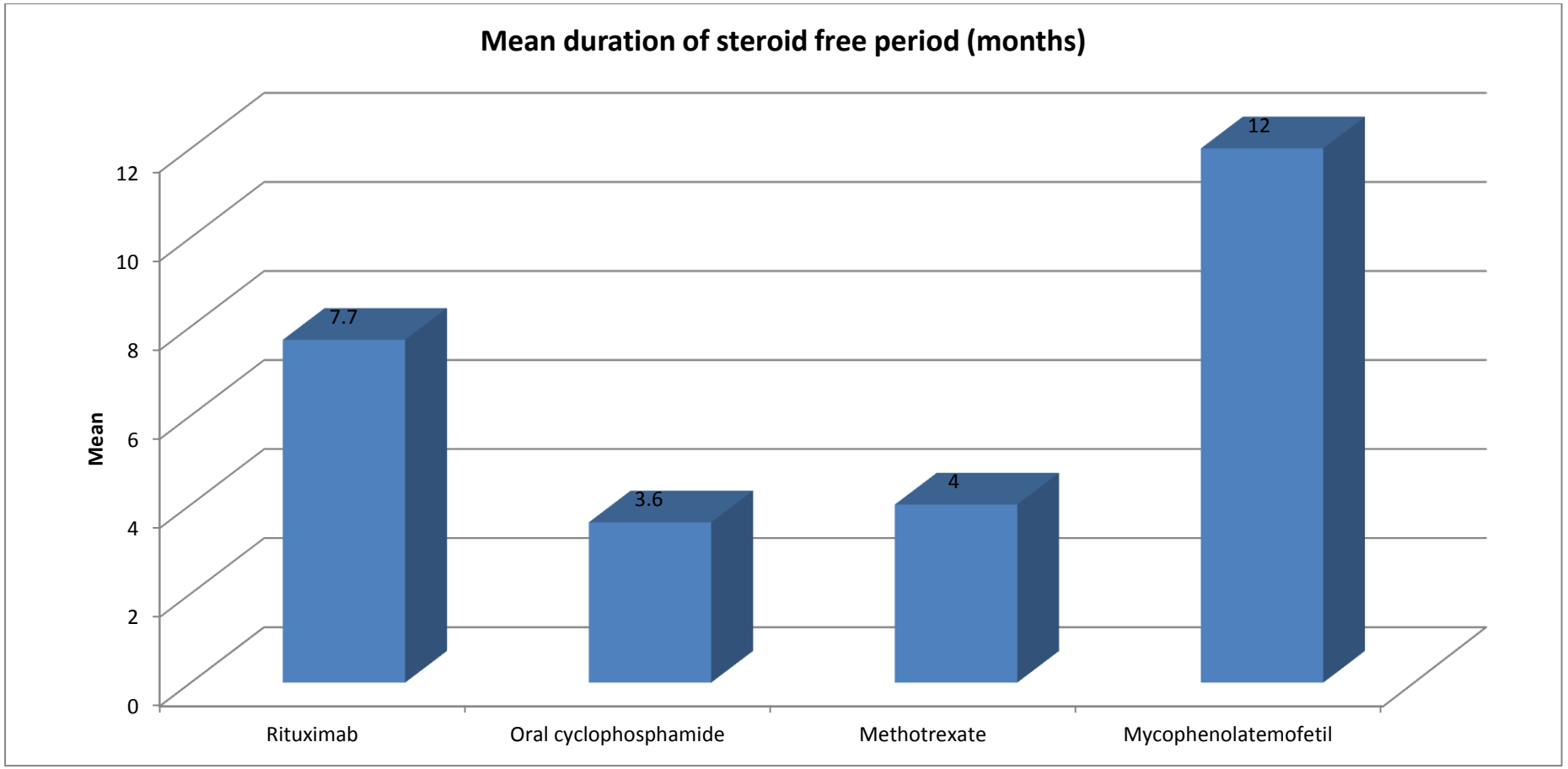

Figure 19: Mean duration of steroid free period with different strategies

e) Table 47: Distribution of patients according to CNI free status

		Strategy implied										Total
		RITUXIMAB		CYCLOPHOSPHAMIDE		MYCOPHENOLATE		METHOTREXATE		DECREASED DOSE OF TACROLIMUS		
	Y	8	42.1%	2	33.3%	1	100.0%	1	50.0%	0	0.0%	12
	N	11	57.9%	4	66.7%	0	0.0%	1	50.0%	1	100.0%	17
Total		19	100.0%	6	100.0%	1	100.0%	2	100.0%	1	100.0%	29

CNI could be stopped in 8 patients given Rituximab, 2 patients given oral cyclophosphamide, 1 patient given MMF, 1 patient given methotrexate for a variable period of time (exact duration in next table).

f) Table 48: Duration of CNI free period in various strategies

	Strategy	N	Mean	SD	median	IQR		Minimum	Maximum
If yes, duration off CNI (in months)	RITUXIMAB	8	3.38	2.97	2.00	1.25	6.00	1.00	9.00
	CYCLOPHOSPHAMIDE	2	10.00	2.83	10.00	8.00	12.00	8.00	12.00
	MYCOPHENOLATE	1	3.00	.	.	.	.	3.00	3.00
	METHOTREXATE	1	9.00	.	9.00	9.00	9.00	9.00	9.00
	DECREASED DOSE OF TACROLIMUS	0	.	.	.	.	.	.	.
	Total	12	4.92	3.82	1.10	2.49	7.35	1.00	12.00

The mean duration of CNI free period was 3.38 months with Rituximab (n=8 patients), 10 months with oral cyclophosphamide (n=2 patients), 3 months with MMF (n=1), 9 months with methotrexate (n=1)

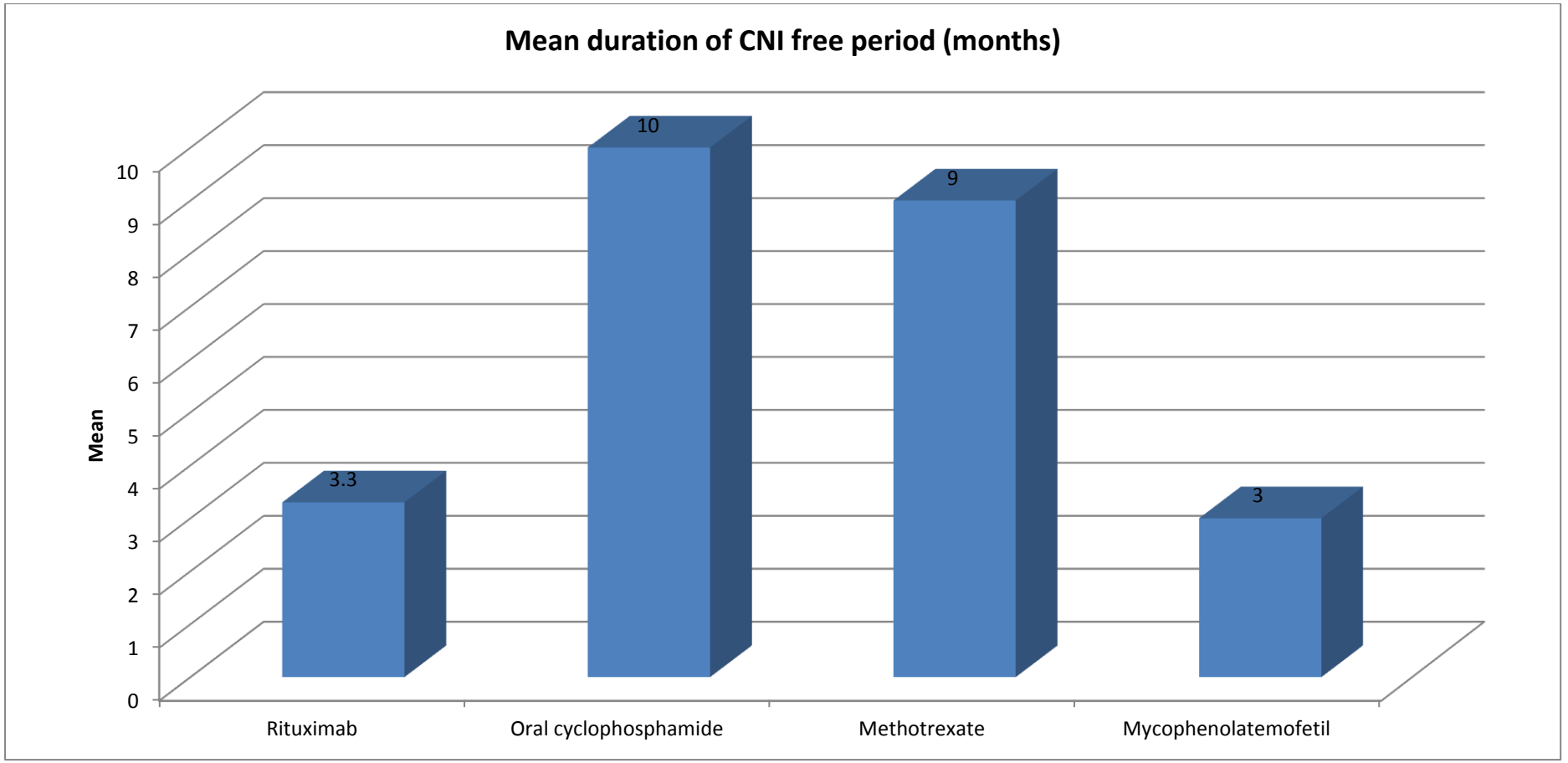

Figure 20: mean duration of CNI free period with each strategy

g) Table 49: Dose of CNI being continued after change in therapy with different strategies

	Strategy	N	Mean	SD	median	IQR		Minimum	Maximum
CNI dose(mg/kg/day) being continued	RITUXIMAB	12	0.07	0.07	0.05	0.04	0.07	0.02	0.25
	CYCLOPHOSPHAMIDE	4	0.08	0.02	0.08	0.07	0.10	0.06	0.10
	MYCOPHENOLATE	0	.	.	3.00	3.00	3.00	.	.
	METHOTREXATE	1	0.07	.	0.07	0.07	0.07	0.07	0.07
	DECREASED DOSE OF TACROLIMUS	1	0.03	.	0.03	0.03	0.03	0.03	0.03
	Total	18	0.07	0.05	0.01	0.04	0.10	0.02	0.25

The mean dose of CNI being continued was 0.07 mg/kg/day with an IQR 0.04-0.1 mg/kg/day

h) Table 50: Number of relapses post 6 months change in therapy

		Strategy employed										Total
		RITUXIMAB		CYCLOPHOSPHAMIDE		MYCOPHENOLATE		METHOTREXATE		DECREASED DOSE OF TACROLIMUS		
No. of relapses after change in therapy (in 6 months)	0	12	63.2%	3	50.0%	1	100.0%	0	0.0%	1	100.0%	17
	1	7	36.8%	2	33.3%	0	0.0%	2	100.0%	0	0.0%	11
	2	0	0.0%	1	16.7%	0	0.0%	0	0.0%	0	0.0%	1
Total		19	100.0%	6	100.0%	1	100.0%	2	100.0%	1	100.0%	29

In the six months period, 12 (63.2%) patients on rituximab, 3(50%) patients on oral cyclophosphamide, 1(100%) patient on MMF and 1 patient (100%), in whom tacrolimus dose was being reduced, had no relapse.

7 (36.8%) patients given Rituximab had 1 relapse each in the six months follow up period. With cyclophosphamide, 2 (33.3%) patients had 1 relapse, 1 patient (16.7%) had 2 relapses.

Patient given MMF had no relapse. Both the patients who were given methotrexate had 1 relapse each.

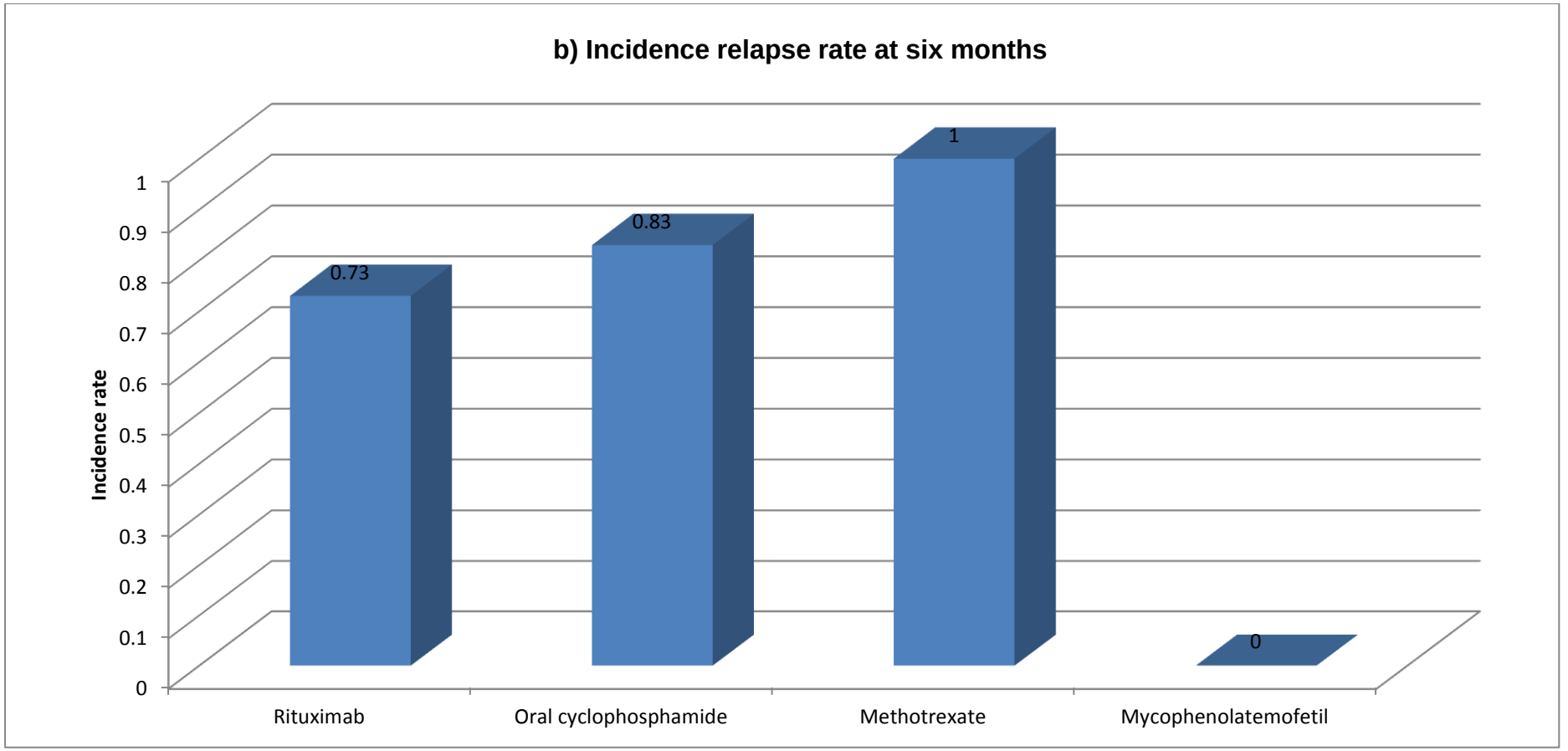

Figure 21: Incidence relapse rate at 6 months with different strategies (expressed as number of relapses/patient years)

i) Table 51: Number of relapses post 12 months change in therapy

		Strategy employed										Total
		RITUXIMAB		CYCLOPHOSPHAMIDE		MYCOPHENOLATE		METHOTREXATE		DECREASED DOSE OF TACROLIMUS		
No. of relapses after change in therapy (in 12 months)	0	6	31.6%	1	16.7%	1	100.0%	0	0.0%	1	100.0%	9
	1	9	47.4%	2	33.3%	0	0.0%	1	50.0%	0	0.0%	12
	2	3	15.8%	0	0.0%	0	0.0%	1	50.0%	0	0.0%	4
	3	0	0.0%	1	16.7%	0	0.0%	0	0.0%	0	0.0%	1
Total		18	94.7%	4	66.7%	1	100.0%	2	100.0%	1	100.0%	26

In the 12 months period, 6 (31.6%) patients on rituximab, 1 (16.7%) patients on oral cyclophosphamide, 1(100%) patient on MMF and 1 patient (100%), in whom tacrolimus dose was being reduced, had no relapse.

9 (47.4%) patients given Rituximab had 1 relapse each whereas 3 (15.8%) patients had 2 relapses in the 12 months follow up period. None of the patient had more than 2 relapses.

With cyclophosphamide, 2 (33.3%) patients had 1 relapse, 1 patient (16.7%) had 3 relapses.

Patient given MMF had no relapse.

Amongst the patients who were given methotrexate, 1 had 1 relapse each while the other had 2 relapses over 12 months.

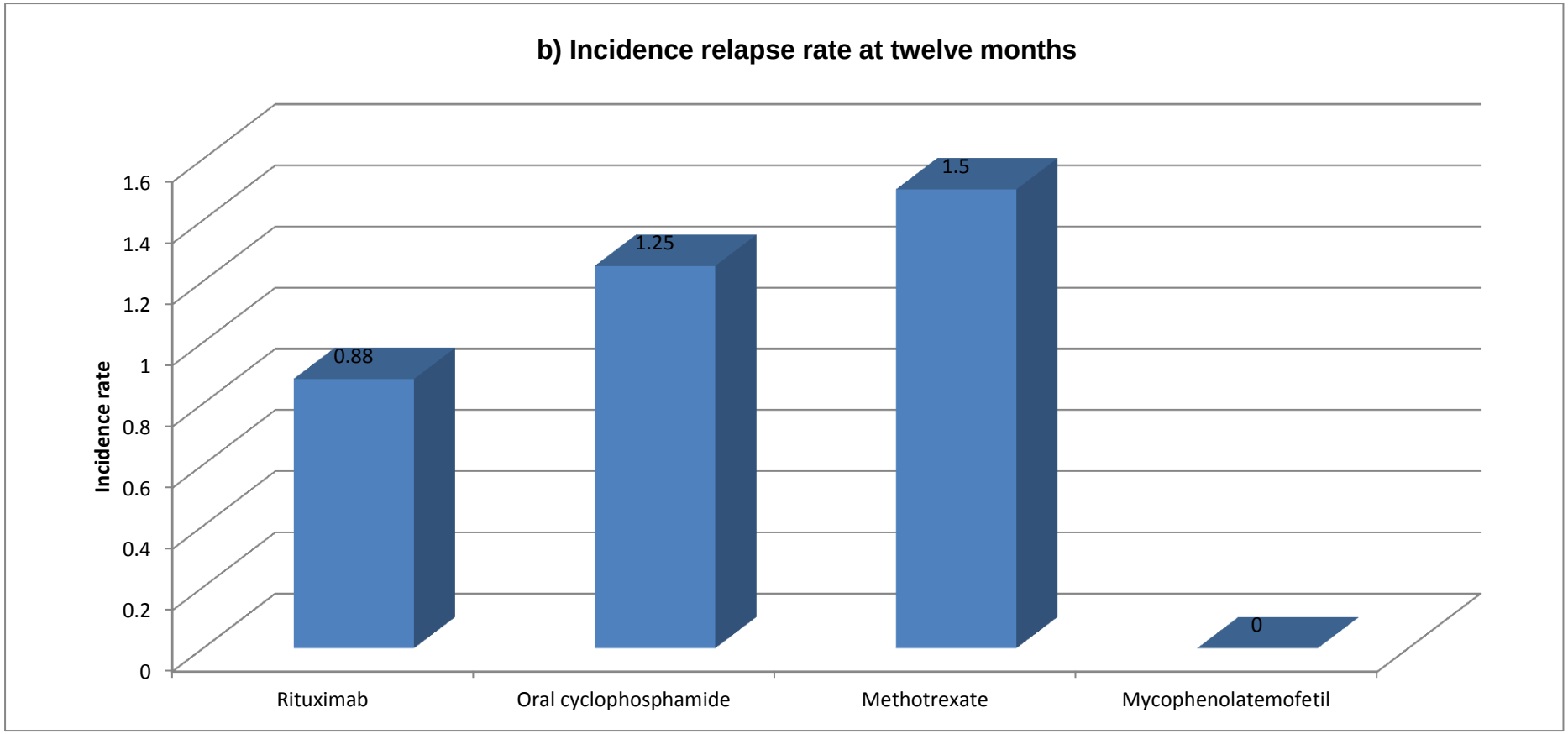

Figure 22: Incidence relapse rate at 12 months with different strategies (expressed as number of relapses/patient years)

j) Table 52: Treatment failure with different strategies

		Strategy implied										Total
		RITUXIMAB		CYCLOPHOSPHAMIDE		MYCOPHENOLATE		METHOTREXATE		DECREASED DOSE OF TACROLIMUS		
Treatment failure(Y/N)	N	19	100.0%	4	66.7%	1	100.0%	1	50.0%	1	100.0%	26
	Y	0	0.0%	2	33.3%	0	0.0%	1	50.0%	0	0.0%	3
Total		19	100.0%	6	100.0%	1	100.0%	2	100.0%	1	100.0%	29

Treatment failure as per the predefined criteria was seen in none of the patients given Rituximab.

It was seen in 2 patients given oral cyclophosphamide. patient no. 23 and 26 , both SDNS, as they showed frequent relapses on cyclophosphamide therapy.

The third patient was with SDNS, who was given methotrexate, therapy failure was in the form of non withdrawal of either steroids or CNI.

29. Table 53: Comparative results of various strategies employed:

Parameter studied	Rituximab	Oral cyclophosphamide	Methotrexate	Mycophenolate mofetil
No. of cases who were prescribed the particular drug	19	6	2	1
SDNS 18	9	5	2	1
SRNS 11	10	1	0	0
Total no. of cases followed for 6 months **(n=29)**	19	6	2	1
Efficacy at 6 months **(n=29)**				
a) in terms of remission				
CR (SDNS/SRNS)	14 (7/7)	4 (3/1)	2 (2/0)	1 (1/0)
PR (SDNS/SRNS)	4 (1/3)	2 (2/0)	0	0
NR (SDNS/SRNS))	1(1/0)	0	0	0
b) Incidence relapse rate (relapses/patient/6 months)	0.73	0.83	1	0
Total no. of cases followed for 12 months **(n=25)**	17	4	2	1
Efficacy at 12 months **(n=25)**	15(8/7)	3(2/1)	2(2/0)	1(1/0)
b)in terms of remission	2(1/1)	0	0	0
CR (SDNS/SRNS)	0	1(1/0)	0	0
PR (SDNS/SRNS)	0.88	1.25	1.5	0
NR (SDNS/SRNS)				
b) Incidence relapse rate (relapses/patient/year)				

No. of patients who were steroid free	19(100%)	6(100%)	1(50%)	1(100%)
Mean steroid free duration (months)	7.7	3.6	4	12
No. of patients in whom CNI could be withdrawn	8 (42.1%) 5 SDNS 3 SRNS	2 (33.3%) 2 SDNS 0 SRNS	1 (50%) SDNS	1 (100%) SDNS
Mean CNI free duration (months)	3.3	10	9	3

A) Rituximab : It was used in 9 patients of SDNS and 10 patients of SRNS (total n=19); 17 patients followed up for 12 months

- 15 of these were in complete remission (8 SDNS, 7 SRNS) on follow up.
- 2 patients was in partial remission (one each of SDNS and SRNS)
- All the patients could be made steroid free (mean duration 7.7 months)
- CNI could be stopped in 42% patients (mean duration 3.3 months)
- 6 patients had no relapse, 9 had 1, and 3 patients had 2 relapses on rituximab therapy
- Incidence relapse rate with Rituximab was 0.88 relapses/patient year

B) Cyclophosphamide : It was given to 5 patients of SDNS and 1 patient of SRNS; 4 followed up for 12 months

- 3 patients were in complete remission (2 SDNS, 1 SRNS) after 12 months
- One patient (SDNS) was in non remission
- Steroids could be withdrawn in all the patients for variable period of time (mean duration 3.6 months)

- CNI could be completely withdrawn in 2 patients (both SDNS).
- In patient with SRNS, dose reduction of CNI was seen by 40% and that patient had no relapse in 12 months period
- Incidence relapse rate with cyclophosphamide was 1.25 relapses/patient year
- Treatment failure was seen in 2 patients ; patient no. 23 and 26 , both SDNS, as they showed frequent relapses on cyclophosphamide therapy.

C) Methotrexate : It was used in 2 patients; both SDNS; both followed up for 12 months

- Both the patients were in complete remission at 12 months follow up.
- One of these could be made steroid free for 4 months and CNI free for 9 months
- Incidence relapse rate with methotrexate was 1.5 relapses/patient year
- Treatment failure was seen in one patient in form of non withdrawal of steroids and CNI (though dose reduction by around 30% was seen)

D) Mycophenolate mofetil : It was given to one patient of SDNS; followed for 12 months

- The patient was in complete remission at the end of 12 months follow up
- There was no relapse in this period
- CNI was gradually tapered off over 9 months, while increasing the dose of MMF

30. Incidence of side effects observed with the use of CNI sparing agents:

Table 54: Incidence of side effects after change in therapy

	SDNS	Percentage	SRNS	Percentage	Total
HYPOGAM-MAGLOBULINEMIA	1	5.6%	0	0.0%	1
LEUCOPENIA	1	5.6%	1	9.1%	2
N	16	88.9%	10	90.9%	26

Transient hypogammagulinemia was seen in one patient with SDNS on rituximab therapy while transient leucopenia was seen in two patients on oral cyclophosphamide therapy (1 SDNS, 1 SRNS). Both these side effects were transient and reversible.

DISCUSSION

The study was done in the Department of Nephrology and Paediatrics in children attending outpatient department of Dayanand Medical College, Ludhiana. Ours was an observational study. It included both prospective and retrospective cases during the study period.

The retrospective cases enrolled were the previously diagnosed cases of SDNS and SRNS, who were being given either of the CNI sparing drugs and now visited OPD for follow up within the study period.

The prospective cases were the SDNS and SRNS patients who were already on calcineurin inhibitors for atleast 12 months before enrolment and were started on any of the CNI sparing strategies for The efficacy of individual drugs as CNI sparing agents has been reported in various studies. However, the studies on different strategies being used for maintenance of remission in SDNS and SRNS in a single centre from India are lacking. This study was done to observe the different CNI sparing strategies being used by the treating clinicians for the long term management of such cases.

Of the children attending Nephrology and Paediatric Nephrology clinic of DMCH, 29 fulfilled the inclusion criteria and were thereby enrolled into the study. 18 of these patients were having steroid dependent nephrotic syndrome and 11 were having steroid resistant nephrotic syndrome. Patients were already on calcineurin inhibitor for a minimum of twelve months and were in either complete (22, 75.9% patients) or partial remission (7, 24.1%patients) at the time of enrolment. They were followed up for 6-12 months period.

In our study, amongst the 29 participants enrolled, 18 (62%) were having steroid dependent nephrotic syndrome and 11 (37.9%) were having steroid resistant nephrotic syndrome. Earlier studies done on alternative therapies also had a higher number of SDNS patients enrolled as compared to SRNS. In the study by Sinha et al, there were 101 patients in SDNS group and 34 in SRNS group.[42]

Ito et al enrolled 53 (74.3%) children with SDNS and 19 (25%) children with SRNS in their study on the use of rituximab as a treatment option for these patients.[56]

The higher number of SDNS in our and these studies could be explained by the overall prevalence of steroid dependent nephrotic syndrome being greater than steroid resistant nephrotic syndrome.

Gender

It was noted that a higher number of participants in our study were male, i.e. 24 (82.8%) children were male and 5 were female, resulting in a male to female ratio of 4.8:1. This distribution is similar to other studies which have shown a male predominance in the prevalence of INS in children.

Sinha et al reported a male predominance in their study on difficult to treat nephrotic syndrome. In their study, 68.3% children were male in the SDNS group and 55.9% in the SRNS group.[42]

Similarly, in a study on the effectiveness of Rituximab in nephrotic syndrome by Prytula et al, 89% in SDNS group and 63% in SRNS group were male children.[57]

Ravani et al too, in their study, reported a male predominance of SDNS patients with 63% study participants being male.[52]

Age distribution

The age at onset of nephrotic syndrome in the study group ranged from six months to 7.5 years. The maximum number of children (89.7%) had disease onset between 1-5 years of age, the mean age being 2.77 years. In a similar study by Prytula et al, the age of onset of nephrotic syndrome ranged from 18 months to 17 years and the mean age was 3.5 years.[57]

The mean age during enrolment i.e. at the beginning of CNI sparing strategy was 7.29 years. This was similar to the randomised control trial conducted by Basu et al where the mean age of study participants was 7.2 years.[58] While in study by Ravani et al, the mean age of patients was 10 years.[52]

In study by Sinha et al, the mean age at nephrotic syndrome onset was 3.3 years and the mean age at CNI sparing strategy (Rituximab) administration was 9.1 years.[42]

Thus, in most of the studies on difficult to treat/ steroid dependent/steroid resistant nephrotic syndrome, a long period between the disease onset and administration of either of the CNI sparing agent exists, as seen in our study.

Associated features

In our study, 10 (34.5%) children had hypertension and 3 (10.3%) had microscopic haematuria seen at the onset of NS, along with other features. The prevalence of hypertension varies widely in various studies. The ISKDC found that 12.8% with MCD had diastolic BP above the 98th percentile. Similarly, 13.4% with focal segmental glomerulosclerosis (FSGS) also exhibited elevated blood pressure.[59]

Steroid related side effects

The most common steroid related side effect observed in our patient population was presence of cushingoid features in 4 (13.8%) patients, followed by cataract in 2 (6.9%) patients. All of the patients exhibiting such features were in the SDNS group as they were on steroids for a long period of time.

In study by Sinha et al, the commonest features of steroid toxicity seen were obesity with BMI >2 SDS and low height (<- 2 SDS) followed by cataract, hypertension, diabetes, etc.[42]

Renal biopsy findings

Kidney biopsy is usually not performed in most patients with INS, esp. steroid sensitive NS. In our study, biopsy was not performed in 19 (65.5%) patients. Amongst the patients in whom biopsy was perfomed, the most common finding was minimal change nephropathy followed by FSGS. Similar to our study, in the study on SDNS by Ravani et al, biopsy was not done in majority i.e. 65% of the patients.[52]

Conversely, in the RITURNS trial conducted by Basu et al in 2016 to study the effect of rituximab vs tacrolimus in SDNS, kidney biopsy was conducted in all patients. The commonest finding on renal biopsy, like our study, was MCD followed by FSGS.[58]

Prior immununosuppression

It was noted that 15 out of 29 children (51.7%) had received other steroid sparing immunosuppressive agents before use of CNIs. Out of these, 4 children (13.7%) had been given three immunosuppressive agents prior to treatment with CNIs.

While either of the two calcineurin inhibitors was the first agent to be used in 14 (48.2%) patients; majority being in the SRNS group.

In earlier studies too, a number of steroid sparing agents had been tried before giving CNIs. All patients in the study by Prytula et al had received two or more treatments prior to Rituximab.[57]

Type and duration of CNI being used

In this study, 25 (86.2%) patients were receiving tacrolimus and 4 (13.8%) were on cyclosporine prior to change in therapy. This was purely on the discretion of treating physician and could be due to the better side effect profile and tolerability noted with tacrolimus in earlier studies.

The mean period of therapy with CNI was 2.9 years. Earlier studies have shown CNI use for variable periods before the introduction of CNI sparing strategy.

Ravani et al enrolled patients only after calcineurin inhibitor had been used for a mean period of 5 ± 3.5 years.[52]

In study by Gellermann et al, sequential therapy with MMF was studied in SRNS, whereby MMF was added to the treatment protocol after initially using cyclosporine for an average 6 months period followed by gradual tapering after addition of MMF.[49]

In study by Sinha et al, all patients of SRNS group had received CNI for a duration of approx. 3.6 ± 1.6 years.[42]

In the latest KDIGO and IPNA guidelines, it is recommended to use CNI therapy for SRNS for a minimum of 1 year in patients who are CNI responsive. Accordingly, in our study, it was observed that CNI was continued for a minimum period of 12 months prior to change in therapy.[2,35]

Drug given

Large scale clinical trials for the management of patients of SDNS and SRNS, after achievement of initial remission with calcineurin inhibitors, are lacking. Thus, management of such cases has been done in an empirical and individualised fashion by the treating physicians. The studies so far have shown variable responses to different therapies in this context.

In our study, it was observed that Rituximab was used in the majority of patients of SDNS (50%) and SRNS (90%) for maintenance of remission following therapy with either of the CNIs. Oral cyclophosphamide was used in 27% of SDNS and only 1 patient (9%) of SRNS. MMF was used in only one patient of SDNS with gradual tapering off of cyclosporine. Methotrexate was used in 2 patients (both SDNS).

Earlier studies have reported the administration of rituximab, MMF and cyclophosphamide as CNI sparing therapies with variable response.

Dose given

The optimal dose of rituximab for the treatment of NS has not been established. Different studies with one, two or more infusions of rituximab at a dose of 375 mg/m^2 each have been done earlier. Kamei et al reported 12 patients of SDNS where a single rituximab dose was used to achieve a high rate of initial response.

Similar to ours, in the study by Sinha et al, the majority of patients (82%) received 2 infusions of Rituximab [42] while in study by Prytula et al, the majority of patients were administered 4 doses of rituximab at weekly intervals.[57]

Cyclophosphamide was given orally at a dose of 2 mg/kg/day for 12 weeks as used in previous studies.[20] MMF was used in a sequential manner with simultaneous tapering of cyclosporine, similar to study by Gellermann et al.[49]

Follow up

Patients were followed up for a variable period ranging from 6 months to 1 year after change in therapy. It was similar to study by SInha et al.[42] While in the study by Kamei et al, the monitoring period for clinical

parameters was only 6 months [43] and in study by Gellerman et al, it was 7 years.

Primary outcome (Sustained remission after change in therapy)

Steroid and relapse free period was more than six months in 16 (i.e. 57.1%) patients.

37% patients with SDNS and 33% patients with SRNS had no relapse in the 12 month period after change in therapy.

SDNS

It was observed that at the end of 12 months, 87.5% patients of SDNS were in CR, 1 (6.25%) was in PR while one patient was not in remission.

Out of these 14 patients who achieved CR, Rituximab was used in 8 patients, cyclophosphamide was used in 2 patients, methotrexate was used in 2 patients, MMF in 1 patient and tacrolimus dose reduction was done in 1 patient.

CNI could be withdrawn in 9 (50%) patients. In 5 of these 9 patients of SDNS, CNI were withdrawn within 6 months of change in therapy. There was no significant difference between the duration of stopping CNI with different strategies.

SRNS

It was observed that at the end of 12 months, 88.8% patients of SRNS were in CR and 1 (11.1%) was in PR.

Out of these 8 patients who achieved CR, Rituximab was used in 7 patients, cyclophosphamide was used in 1 patients.

CNI could be stopped in only 3 (27.3%) patients. CNI could be withdrawn in only 1 patient within 6 months of change in therapy.

Response with Rituximab

With the use of Rituximab as CNI sparing therapy 73.7% patients were in CR, 21% were in PR at 6 months. While 88.2% patients were in CR and 11.7% were in PR at 12 months follow up.

Thus, the use of Rituximab showed a favourable response in the majority of patients except one patient each of SDNS and SRNS. Both

these patients were in CR at enrolment, in CR at 6 months after Rituximab but in PR at 12 months. This was due to relapse after 6 months of Rituximab. They showed infrequent relapses in the 12 month period and CNI dose reduction was seen by around 30%.

Similar positive response was seen in the study by Sinha et al using 2-4 weekly doses of Rituximab. In their study, sustained remission was seen in 85% in SDNS and 33.3% in SRNS group at 12 months. Our study showed a better response in SRNS patients managed with Rituximab in terms of remission.

We observed that all patients remained steroid free (mean duration 7 months) and CNI could be stopped in 42% patients (mean duration 3.3 months).

In study by Sinha et al [42], prednisolone was discontinued in 70-90 % patients after a mean period of about 5 months and CNI was discontinued in around 90% of patients after Rituximab therapy (higher than our study)

Ito et al in their study on 53 patients of SDNS and 19 patients of SRNS were able to withdraw steroids in 77% SDNS and 41% SRNS respectively after Rituximab treatment.[56] They could withdraw Cyclosporine A in 60 % of SDNS patients but in none of the patients suffering from SRNS. In contrast, in our study, CNI was withdrawn in 55% of SDNS and 37.5 % of SRNS patients.

Response with oral cyclophosphamide

Oral cyclophosphamide was used as CNI sparing agent in 5 SDNS and 1 SRNS patient. CR was seen in 66.6%, PR in 2 (33.3%) patients at 6 months; and on follow up period of 12 months, 75% showed CR.

One patient given oral cyclophosphamide who was in CR at enrolment, had 3 relapses post change in therapy and was in non remission at 12 months follow up. Tacrolimus in this patient could not be tapered off. He was a difficult to treat NS and had also been given levamisole and MMF before CNI. He was on oral tacrolimus for 3 years and had developed features of steroid toxicity (cataract) and hypertension on tacrolimus. Rituximab remains an option in this patient, which may show better results.

Though cyclophosphamide has been studied as steroid sparing agent earlier, there has been no major study on the efficacy of oral cyclophosphamide as CNI sparing agent.

Sandhu et al in 2021 studied the effect of oral cyclophosphamide in 100 patients with SDNS and reported a response rate of 50% with relapse free survival of 31% at 1 year, 11% at 2 years.[61]

The mean steroid free duration was 3.6 months and CNI was withdrawn in 50 % patients (mean duration 10 months). Thus oral cyclophosphamide could be considered an effective management strategy in maintaining remission in SDNS patients. As it was used in only one patient of SRNS, more data are needed to show its efficacy in this population.

Response with MMF

MMF was used in one patient of SDNS in our study; he continued to maintain sustained remission without any relapse and cyclosporine was gradually tapered off after 9 months.

There have been few studies showing mixed results with use of MMF as a CNI sparing agent. Fujinaga et al in 2007 studied MMF as CNI sparing agent in 12 patients with SDNS and showed that cyclosporine could be weaned off in 9 (75%) patients.[50] The number of relapses on MMF therapy decreased in all patients significantly.

While Gellermann et al studied sequential maintenance therapy with cyclosporine A and MMF showing that this protocol maintained remission in all patients during a follow up time of 7.0 years.[49] They were able to maintain 13 (57%) patients on MMF monotherapy. Likewise, we too maintained our patient on MMF monotherapy and he is doing well so far.

MMF has earlier been studied as an agent for initial induction of remission but failed to show positive results. It can thus be incorporated as an effective agent to maintain remission after CNI therapy in these patients.[60]

Response with Methotrexate

It was given to 2 patients. At 12 months, both patients were in CR but steroids and CNI could not be tapered off in 1 of them. There have

been limited studies on use of methotrexate in SDNS patients; it has been used in patients having RA with NS but our patients didn't have any features of arthritis. The use of methotrexate was solely on the discretion of the treating physician, before widespread use of Rituximab was available.

Relapse rate after change in therapy

Time to first relapse

In our study, it was observed that the mean time to first relapse was 5.6 months (IQR 4-7.7) after change in therapy. There was no significant difference between different strategies.

In study by Sinha et al, the median time to relapse in children who completed steroid and CNI treatment was 5.6 to 7.3 months after the first rituximab infusion and 8.5 - 11.6 months after the second and subsequent infusions.[42]

Incidence relapse rate after therapy change

The incidence relapse rate was 1.55 on CNI therapy, 0.48 post 6 months of change in therapy and 0.88 post 12 months of change in therapy (significant decline, p =0.005)

In study by Fujinaga et al, number of relapses decreased significantly after a single infusion of Rituximab (p 0.02).[50] In study by Sinha et al, relapse rate in SRNS patients showed a significant decline by 71% ($p<0.0001$).[42]

Effect on eGFR

eGFR improved after switching therapy from CNI to other agents with a significant (p=0.03) increase in SDNS and insignificant increase in SRNS (0.261).

Side effects with new drug

We did not observe severe side effects with any of the new strategy employed. Transient hypogammagulinemia was seen in one patient on rituximab therapy while transient leucopenia was seen in two patients on oral cyclophosphamide therapy.

Risks associated with discomfort during rituximab infusion can be minimized with pre treatment and by giving the infusion at a slow rate. All these precautions were followed in our study.

In the study by Fujinaga et al, mild reactions to Rituximab infusions were seen in around 50% of the patients, but major side effect like severe infections requiring hospitalisation were not seen in any of the patient.

Kamei et al reported high incidence (36%) of severe respiratory injuries associated with the use of rituximab, such as interstitial pneumonitis, acute respiratory distress syndrome, or lung fibrosis.

Treatment failure

Treatment failure was observed in 3 patients. 2 patients given oral cyclophosphamide. patient no. 23 and 26 , both SDNS, as they showed frequent relapses on cyclophosphamide therapy.

The third patient was with SDNS, who was given methotrexate, therapy failure was in the form of non withdrawal of either steroids or CNI.

Limitations

A major limitation was smaller number of patients in our study. As the data were both retrospective and prospective, investigation records including CD 19 levels were not available for some patients. It was an observational study where treatment of SDNS and SRNS was as per the discretion of the treating physician and thus varied between the patients. Taking into consideration that prevalence of SDNS and SRNS is not that common, and this study being a single centre study, these limitations are not completely unavoidable.

SUMMARY AND CONCLUSION

SUMMARY

- The present study was done in Nephrology and Paediatrics OPD of a tertiary care hospital, DMCH Ludhiana on patients between 6 months and 18 years of age who were having SDNS or SRNS and were in remission with calcineurin inhibitors. The efficacy of various CNI sparing strategies in this study population was observed.
- Total 29 patients were included in the study. 18(62%) belonged to SDNS and 11 (37.9%) belonged to SRNS category.
- There were 16 children from urban and 13 children from rural background.
- In our study, there were 24 (82.8%) male children and 5 female children thereby showing male predominance.
- The maximum number of children (89.7%) had disease onset between 1-5 years of age, the mean age being 2.77 years.
- Out of all patients, 10 (34.5%) patients had hypertension and 3 (10.3%) had microscopic hematuria at the onset of nephrotic syndrome
- Kidney biopsy was not done in 19 (65.5%) patients. It was done in 10 patients; 6 (20.7%) were diagnosed as having minimal change disease, 3 (10.3%) had FSGS and 1(3.4%) had podocytopathy (not specified, electron microscopy not done).
- Out of 29 patients, steroids were being given in 13 patients at the time of enrolment into our study. All these patients were having SDNS.
- In 15 (51.7%) patients, patients had been given other immunosuppressants prior to using calcineurin inhibitors. MMF was the most frequently used agent in SDNS (used in 44.4%) and cyclophosphamide was the most common agent used in SRNS patients (used in 45.5%). In 14 (48.2%) patients, calcineurin inhibitors were the first agents to be used.

- 25 (86.2%) children were on tacrolimus and 4 (13.8%) were on cyclosporine at the time of enrolment.
- The mean duration of CNI use was 2.9 years prior to initiation of CNI sparing strategies.
- 22 (75.9%) patients were in complete remission and 7 (24.1%) were in partial remission at the time of enrolment.
- The mean age of the patients at the beginning of study i.e. when CNI sparing agent was started, was 7.29 (IQR 5.5-9.3) years.
- Rituximab was the most common CNI sparing agent used (65.5%) followed by oral cyclophosphamide (20.7%), methotrexate (6.9%) and MMF (3.4%).
- The mean time to first relapse was 5.6 months (IQR 4-7.7) after change in therapy
- The incidence relapse rate was 1.55 on CNI therapy, 0.48 post 6 months of change in therapy and 0.88 post 12 months of change in therapy (significant decline, p = 0.005)
- eGFR improved after switching therapy from CNI to other agents with a significant (p=0.03) increase seen in SDNS patients and insignificant increase in SRNS (0.261).
- There were no major side effects observed with any of the strategy used in our study
- Strategy wise response rate:

a) **Rituximab** :
   - Used in 19 patients (9 SDNS, 10 SRNS)
   - CR was seen in 88.2% patients (53.3% SDNS, 46.6% SRNS) and PR in 11.7% (50% SDNS, 50% SRNS) at 12 months.
   - 1 SDNS patient who was earlier in CR went into PR at 12 months
   - All the patients were steroid free (mean duration 7.7 months)

- CNI could be stopped in 42% patients (mean duration 3.3 months); 5 SDNS and 3 SRNS
- Incidence relapse rate with Rituximab was 0.88 relapses/patient year.

b) Oral Cyclophosphamide :

- It was given to 6 patients (5 SDNS, 1 SRNS)
- CR was seen in 75% patients (66% SDNS, 33.3% SRNS), non remission in 25% (1 SDNS patient) at the end of 12 months follow up.
- The mean steroid free duration was 3.6 months
- CNI were withdrawn in 33.3 % patients (mean duration 10 months); both SDNS
- Incidence relapse rate was 1.25 relapses/patient year
- Treatment failure was seen in 2 patients ; both SDNS (showed frequent relapses).

c) Methotrexate :

- It was used in 2 patients (both SDNS)
- CR was seen in both at 12 months
- One of these could be made steroid free for 4 months and CNI free for 9 months
- Incidence relapse rate with methotrexate was 1.5 relapses/patient year
- Treatment failure was seen in one patient in form of non withdrawal of steroids and CNI (though dose reduction by around 30% was seen)

d) MMF

- The only patient of SDNS given MMF was in CR at the end of 12 months.
- There was no relapse in this period
- CNI was gradually tapered off over 9 months

CONCLUSION

The present study was conducted to observe the various CNI sparing strategies used in SDNS and SRNS. Rituximab was the most frequently used agent followed by oral cyclophosphamide.

Our study confirmed that Rituximab was effective in maintaining sustained remission in both SDNS and SRNS category and reduced the steroid and CNI burden to a considerable extent.

Whereas, use of oral cyclophosphamide as CNI sparing agents showed mixed response. CNI could not be tapered off in the only patient of SRNS given oral cyclophosphamide

Methotrexate showed a variable and MMF showed a good clinical and immunological response but the number of patients who were prescribed these drugs was very small.

The statistically significant decline in incidence relapse rate and an improvement in eGFR seen in our study adds to the existing data, suggesting that these strategies could be effective for maintaining sustained remission in patients who are on prolonged therapy with calcineurin inhibitors.

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S.No.	NAME	SEX (M/F)	Urban/ rural	Date of birth	Age Of Onset (years)	Diagnosis (FRNS/ SDNS/ SRNS)	Hypertension at onset (Y/N)	Haematuria at onset (Y/N)	Steroid Dependent Dose (mg/kg/day) (If being given)	Prior Immunosuppressives:			Cataract
										Levamisole (Y/N)	Cyclophosphamide (Y/N)	Mycophenolate (Y/N)	
1	HARKIRAT	M	URBAN	24.09.2010	3	SDNS	Y	N	1.5	Y	Y	Y	N
2	SAHIBJOT	M	RURAL	03.01.2015	1.08	SRNS	N	Y		N	N	N	N
3	DIVYA	F	RURAL	19.09.2013	1.41	SDNS	N	N	1	N	N	N	N
4	MUNTAZ	M	RURAL	04.11.2014	1.41	SDNS	Y	N	2	N	N	N	N
5	JASHANPREET	M	RURAL	30.06.2014	4	SRNS	Y	Y		N	N	N	N
6	PRABHNOOR	M	URBAN	05.02.2014	3	SRNS	N	N		N	Y	N	N
7	GURLEEN	F	URBAN	22.04.2010	5	SRNS	N	N		Y	Y	Y	N
8	HARSHDEEP	F	URBAN	30.12.2012	3.5	SDNS	N	Y	1.5	Y	N	N	N
9	MANPREET	M	URBAN	19.08.2013	5	SRNS	Y	N		N	N	N	N
10	KAHNOORPREET	M	RURAL	27.03.2015	4	SDNS	N	N	1	N	N	N	N
11	SUKHMAN	M	URBAN	21.01.2018	2.6	SRNS	N	N		N	Y	N	N
12	KARAMVEER	M	URBAN	06.10.2012	7.5	SDNS	N	N		N	N	N	N
13	ARMAAN	M	RURAL	04.11.2018	1	SDNS	N	N	1	N	N	Y	N
14	RAVJOT	M	RURAL	18.11.2012	1.16	SDNS	Y	N	1.3	N	N	N	N
15	SAURAV	M	URBAN	26.05.2012	1.91	SDNS	N	N	1.25	Y	Y	Y	N
16	SAKSHAM	M	URBAN	28.09.2014	1.5	SDNS	Y	N	2	N	N	N	N
17	KANAV	M	URBAN	13.10.2014	1.6	SRNS	Y	N		N	N	N	N
18	HARJOT	M	RURAL	12.03.2019	0.6	SDNS	N	N		N	N	N	N
19	EKAMJEET	M	RURAL	29.12.2015	3.66	SDNS	N	N		N	N	Y	N
20	KESHAV	M	URBAN	31.07.2016	3	SRNS	N	N		N	N	N	N
21	SAKSHAM	M	URBAN	28.03.2010	2	SDNS	N	N		Y	Y	Y	N
22	INDERJIT	M	RURAL	29.11.2007	0.8	SDNS	N	N		N	Y	Y	N
23	RAGHAV	M	URBAN	06.11.2018	1.5	SDNS	Y	N	0.2	N	N	N	N
24	ARMANDEEP	M	URBAN	07.10.2014	5	SDNS	N	N	0.5	Y	N	N	Y
25	PRIYA	F	RURAL	15.10.2015	4	SRNS	N	N		N	N	N	N
26	KARAN	M	RURAL	10.11.2015	3	SDNS	N	N	1	Y	N	Y	Y
27	DIKSHANT	M	URBAN	24.09.2017	2.08	SDNS	N	N	0.5	N	N	Y	N
28	HEMANT	M	URBAN	14.03.2012	4	SRNS	Y	N		N	Y	N	N
29	SUKHDEEP	F	RURAL	10.10.2016	2	SRNS	Y	N		N	Y	N	N

S.No.	Features of steroid toxicity (Y/N)				Renal biopsy findings if done	CNI being given (tacrolimus/ cyclosprine)	Date of starting CNI	CNI dose being given at start of study (mg/kg/day)	No of Relapses in last One Year (on CNI)	Hypertension
	Cushingoid features	Short stature	Overweight	Obesity						
1	N	N	N	N	PODOCYTOPATHY	TACROLIMUS	29.05.2019	0.12	2	N
2	N	N	N	N	NOT DONE	CYCLOSPORINE	22.04.2016	6.42	0	N
3	N	N	N	N	MCD	TACROLIMUS	05.06.2017	0.23	2	Y
4	N	N	N	N	FSGS	TACROLIMUS	30.11.2016	0.04	2	N
5	Y	N	N	Y	NOT DONE	TACROLIMUS	27.07.2018	0.05	1	Y
6	Y	N	Y	N	MCD	CYCLOSPORINE	13.09.2017	0.1	2	Y
7	N	N	N	N	FSGS	TACROLIMUS	08.08.2020	0.1	3	Y
8	Y	Y	N	N	NOT DONE	TACROLIMUS	30.10.2017	0.07	2	N
9	N	N	N	N	NOT DONE	TACROLIMUS	29.06.2020	0.25	2	N
10	N	N	N	N	NOT DONE	TACROLIMUS	18.09.2020	0.14	2	N
11	N	N	N	N	NOT DONE	TACROLIMUS	10.11.2020	0.15	1	Y
12	N	N	N	N	NOT DONE	TACROLIMUS	24.07.2020	0.17	1	N
13	N	N	N	N	NOT DONE	TACROLIMUS	10.02.2020	0.2	3	N
14	N	N	N	N	MCD	CYCLOSPORINE	01.09.2014	4	1	N
15	N	N	N	N	NOT DONE	TACROLIMUS	25.10.2021	0.1	4	N
16	N	N	N	N	NOT DONE	TACROLIMUS	28.08.2017	0.1	1	N
17	N	N	N	N	NOT DONE	TACROLIMUS	15.11.2017	0.05	2	N
18	N	N	N	N	NOT DONE	TACROLIMUS	07.08.2020	0.09	0	N
19	N	N	N	N	NOT DONE	TACROLIMUS	03.01.2022	0.08	0	N
20	N	N	N	N	MCD	TACROLIMUS	14.11.2019	0.06	3	N
21	N	N	N	N	NOT DONE	TACROLIMUS	14.10.2015	0.09	1	N
22	N	N	N	N	MCD	TACROLIMUS	22.10.2014	0.1	1	N
23	N	N	N	N	NOT DONE	TACROLIMUS	27.07.2020	0.19	1	N
24	Y	N	N	N	NOT DONE	CYCLOSPORINE	06.12.21	4.34	0	N
25	N	N	N	N	FSGS	TACROLIMUS	04.04.2020	0.15	2	N
26	N	N	N	N	NOT DONE	TACROLIMUS	14.06.2019	0.11	1	Y
27	N	N	N	N	NOT DONE	TACROLIMUS	12.08.2020	0.1	1	N
28	N	N	N	N	NOT DONE	TACROLIMUS	11.07.2019	0.06	2	N
29	N	N	N	N	MCD	TACROLIMUS	14.03.2019	0.1	2	N

S.No.	Side effects of CNI (Y/N)													Date of start of study	Status of patients at start of study (CR/PR)
	Hyperkalemia	Seizure	Increased blood sugars	Increase OT/PT	Diarrhoea	UTI	Reversible nephropathy	Hypertrichosis	Gingival hyperplasia	Acne	Skin infections	Major infecion requiring hospitalisation	Others		
1	N	N	N	N	N	N	N	N	N	N	N	N	N	01.10.2022	CR
2	Y	N	N	N	N	N	N	N	N	N	N	N	N	27.07.2019	CR
3	N	N	N	N	N	N	Y	N	N	N	N	N	N	26.10.2020	CR
4	N	N	N	N	N	N	N	N	N	N	N	N	N	26.11.2019	CR
5	N	N	N	N	N	Y	N	N	N	N	N	N	N	04.02.2020	CR
6	N	N	N	N	N	N	N	N	N	N	N	N	N	03.04.2021	PR
7	N	N	N	N	N	N	N	N	N	N	N	N	N	22.07.2022	PR
8	N	N	N	N	N	N	N	N	N	N	N	N	N	26.11.2019	CR
9	N	N	N	N	N	N	N	N	N	N	N	N	N	18.11.2023	PR
10	N	N	N	N	N	N	N	N	N	N	N	N	N	10.02.23	CR
11	N	N	N	N	Y	N	N	N	N	N	N	N	N	21.10.2023	PR
12	N	N	N	N	Y	N	N	N	N	N	N	Y	N	08.08.2023	CR
13	N	N	N	N	N	N	N	N	N	N	N	N	N	20.04.2023	CR
14	N	N	N	N	N	N	N	N	N	N	N	N	N	29.09.2017	CR
15	N	N	N	N	N	N	N	N	N	N	N	N	N	24.04.2023	CR
16	N	N	N	N	N	N	N	N	N	N	N	N	N	19.09.2020	PR
17	N	N	N	N	N	N	N	N	N	N	N	N	N	09.02.2022	CR
18	N	N	N	N	N	N	N	N	N	N	Y	N	N	05.02.2024	PR
19	N	N	N	N	N	N	N	N	N	N	N	N	N	21.06.2023	CR
20	N	N	N	N	N	N	N	N	N	N	N	N	N	05.07.23	CR
21	N	N	N	N	N	N	N	N	N	N	N	N	N	10.08.2018	CR
22	N	N	N	N	N	N	N	N	N	N	N	N	N	09.02.2018	CR
23	N	N	N	N	N	N	N	N	N	N	N	N	N	18.02.24	CR
24	N	N	N	N	N	N	N	N	N	N	N	N	N	04.05.2023	CR
25	N	N	N	N	N	N	N	N	N	N	Y	N	N	05.05.2023	CR
26	N	N	N	N	N	N	N	N	N	N	N	N	N	12.11.2022	CR
27	N	N	N	N	N	N	N	N	N	N	N	N	N	09.06.2023	CR
28	N	N	N	Y	N	N	N	N	N	N	N	N	N	16.01.2023	CR
29	N	N	N	N	N	N	N	N	N	N	N	N	N	20.06.2022	PR

S.No.	Age at start of study (years)	DURATION OF CNI THERAPY (years)	Weight (kgs)	Height (cms)	BMI(kg/m2) in patients more than 5 years age	Baseline investigations									Drug levels (if done)
						Hb(gm/dl)	TLC (/mm3)	Platelet count/mm3	Urine PCR	S. albumin (g/dl)	S.Creat (mg/dl)	eGFR	SGOT	SGPT	
1	12	3.33	40	144	20.4	13.8	12300	342000	0.2	3.6	0.4	149	24	24	
2	4.5	3.25	16	100		10.6	14400	651000	0.1	3.8	0.6	69	22	23	184
3	7.08	3.33	18	103	18	11.4	6100	330000	0.19	3.5	0.8	53	18	16	5.6
4	5	2.91	18	102	17.3	14.1	10100	526000	0.3	3.6	0.4	105	21	27	
5	5.58	2	28	106	25	12.4	7400	225000	0.26	4.26	0.26	168	35	19	
6	7.41	3.91	22	124	15.06	12.4	10600	232000	0.6	4.32	0.34	151	31	10	4.02
7	10.25	1.91	35	129	21.08	12	9000	212000	1.5	3.6	0.4	133	23	26	4.4
8	6.8	1.75	20.4	102	19.61	12.2	8500	414000	0.23	3.8	0.6	70	30	14	
9	10.16	3.08	28.7	133.6	16.3	15	11000	185000	6.15	3	0.4	138	24	20	4.65
10	7.8	2.16	21	117	15.4	11	7600	212000	0.12	3.9	0.5	97	12	18	
11	5.75	2.75	18.3	102	17.59	11.6	7300	205000	1	3.48	0.7	60	16	14	5.21
12	10.8	3	23.2	123	15.23	11	6700	230000	0.1	4	0.8	63	15	20	
13	4.41	3.5	19.7	113	15.5	12	9500	324000	0.1	4.38	0.6	78	25	14	
14	4.83	2.91	22.7	114	17.5	11.8	5500	231000	0.2	3.9	0.5	94	23	21	178
15	10.8	1.75	32.6	134	18.2	12.3	6600	341000	0.23	4.1	0.7	79	24	20	
16	5.91	2.91	20	108	17.2	11	5700	267000	1.3	3.3	0.5	89	17	12	
17	4.25	1.6	17.1	97.5	18.08	12.1	10800	500000	0.2	3.7	0.2	201	18	28	
18	4.41	4	23	112	18.4	11.2	10320	461000	0.3	3	0.39	119	22	13	
19	7.41	1.58	25.4	124	16.6	10.9	9600	326000	0.13	3	0.75	68	44	32	
20	7	3.58	40	128	24.5	12.7	12500	358000	0.2	4.8	0.4	132	32	32	
21	8.3	2.75	25	122	16.89	12.4	7400	236000	0.2	4.1	0.3	168	16	18	
22	10.16	3.75	32	136	17.39	11.6	5600	213000	0.1	4.2	0.3	187	18	20	
23	5.75	4	17.1	100	17.1	10.9	8400	196000	0.2	3.7	0.23	180	15	11	
24	8.5	1.58	35	124	22.8	12.8	7200	205000	0.1	4	0.3	171	14	18	
25	7.5	4.75	26	118	18.7	13.2	7400	244000	0.4	3.8	0.2	244	19	17	
26	7	3.08	22	114	17.05	11.6	8300	416000	0.1	4.2	0.2	235	12	14	
27	5.6	2.91	21	110	17.35	11.2	6500	276000	0.18	4.12	0.2	227	19	20	
28	10.8	4.08	40	130	23.6	12.1	6000	202000	0.2	4.1	0.3	179	14	15	
29	5.66	3.41	19.5	106	17.4	11.5	9900	312000	0.4	3	1.2	112	18	15	

S.No.	Strategy implied	Dose of new drug given	Associated medications	Speicific drug related side effect	Followed up for (months)	Urine PCR at 3 months	Serum albumin at 3 months	Serum creatinine at 3 months
1	RITUXIMAB	375 MG/M2 (2 DOSES)		N	12	0.1	3.9	0.4
2	RITUXIMAB	375 MG/M2 (3 DOSES)	ENALAPRIL	N	12	0.12	3.9	0.5
3	RITUXIMAB	375 MG/M2 (3 DOSES)		HYPOGAMMAGLOBULINEMIA	12	0.14	3.5	0.5
4	RITUXIMAB	375 MG/M2 (3 DOSES)		N	12	0.15	3.8	0.4
5	RITUXIMAB	375 MG/M2 (3 DOSES)	ENALAPRIL	N	12	0.2	4.1	0.3
6	RITUXIMAB	375 MG/M2 (3 DOSES)		N	12	0.2	3.6	0.2
7	CYCLOPHOSPHAMIDE	50 MG ONCE A DAY FOR 3 MONTHS	RAMIPRIL	LEUCOPENIA	12	0.4	3.6	0.4
8	RITUXIMAB	375 MG/M2 (2 DOSES)			12	0.8	3.7	0.16
9	RITUXIMAB	375 MG/M2 (3 DOSES)	RAMIPRIL		7	0.1	3.9	0.4
10	METHOTREXATE	10 MG WEEKLY	ENALAPRIL		12	1.5	3.57	0.4
11	RITUXIMAB	375 MG/M2 (2 DOSES)	ENALAPRIL, AMLODIPINE, THYRONORM		8	0.9	3.3	0.6
12	RITUXIMAB	375 MG/M2 (3 DOSES)			12	0.1	4.1	0.5
13	CYCLOPHOSPHAMIDE	50 MG ONCE A DAY FOR 3 MONTHS			12	0.3	3.8	0.45
14	CYCLOPHOSPHAMIDE	50 MG ONCE A DAY FOR 3 MONTHS	ENALAPRIL		12	0.1	4	0.5
15	METHOTREXATE	10 MG WEEKLY			12	0.3	3.5	0.5
16	RITUXIMAB	375 MG/M2 (2 DOSES)	ENALAPRIL		12	0.8	3.7	0.4
17	RITUXIMAB	375 MG/M2 (4 DOSES)	ENALAPRIL, THYRONORM		12	0.3	4.1	0.2
18	CYCLOPHOSPHAMIDE	50 MG ONCE A DAY FOR 3 MONTHS		LEUCOPENIA	6	0.4	3.4	0.3
19	DECREASED DOSE OF TACROLIMUS				12	0.2	4.1	0.2
20	RITUXIMAB	375 MG/M2 (3 DOSES)	ENALAPRIL		12	0.1	4	0.4
21	RITUXIMAB	375 MG/M2 (3 DOSES)	ENALAPRIL		12	0.1	3.9	0.2
22	RITUXIMAB	375 MG/M2 (3 DOSES)			12	0.1	4.2	0.18
23	CYCLOPHOSPHAMIDE	50 MG ONCE A DAY FOR 3 MONTHS			6	3.5	2.3	0.3
24	MYCOPHENOLATE	1200 MG/M2/DAY			12	0.12	4.2	0.33
25	RITUXIMAB	375 MG/M2 (2 DOSES)	RAMIPRIL		12	0.16	3.7	0.2
26	CYCLOPHOSPHAMIDE	50 MG ONCE A DAY FOR 3 MONTHS			12	0.12	4.1	0.16
27	RITUXIMAB	375 MG/M2 (3 DOSES)	ENALAPRIL		12	0.15	4	0.2
28	RITUXIMAB	375 MG/M2 (3 DOSES)	ENALAPRIL		12	0.1	4	0.2
29	RITUXIMAB	375 MG/M2 (3 DOSES)	ENALAPRIL		12	0.3	3.8	0.4

S.No.	Urine PCR at 6 months	Serum albumin at 6 months	Serum creatinine at 6 months	eGFR at 6 months	Status of patients at 6 months post change in therapy (CR/PR/NR)	Urine PCR at 9 months	Serum albumin at 9 months	Serum creatinine at 9 months	Urine PCR at 12 months	Serum albumin at 12 months	Serum creatinine at 12 months	eGFR at 12 months	Status of patients at 12 months post change in therapy (CR/PR/NR)	Off steroids (Y/N)
1	0.2	3.8	0.45	132	CR	0.8	3.3	0.3	0.12	3.7	0.3	198	CR	Y
2	0.12	3.9	0.6	69	CR	0.1	4.1	0.5	0.08	4.1	0.5	83	CR	Y
3	0.1	3.5	0.5	85	CR	0.1	3.6	0.3	0.12	3.9	0.3	142	CR	Y
4	3	2.5	0.5	84	NR	0.2	3.6	0.45	0.12	3.65	0.31	136	CR	Y
5	0.6	4.26	0.2	219	CR	0.72	3.9	0.4	1.7	3.86	0.26	168	PR	Y
6	0.3	3.5	0.3	171	PR	0.2	3.9	0.2	0.1	4	0.3	171	CR	Y
7	0.2	3.7	0.45	118	CR	0.1	3.9	0.3	0.2	3.6	0.26	205	CR	Y
8	0.2	3.9	0.5	84	CR	0.3	3.5	0.14	2.3	3	0.2	211	PR	Y
9	7.5	2.5	0.3	184	PR									Y
10	0.24	3.6	0.5	97	CR	0.1	3.9	0.45	0.12	3.8	0.4	121	CR	Y
11	0.2	3.93	0.66	64	CR									Y
12	0.02	4.2	0.4	127	CR	0.1	4.1	0.3	0.05	4	0.4	127	CR	Y
13	0.1	4.38	0.51	92	CR	0.4	4	0.43	0.1	4.1	0.4	117	CR	Y
14	0.16	4.1	0.6	78	CR	3.8	2.4	0.5	0.2	3.9	0.4	118	CR	Y
15	0.2	3.6	0.4	138	CR	0.1	4	0.1	0.2	3.9	0.4	138	CR	N
16	1.2	3	0.5	89	PR	3.5	2.2	0.4	0.3	4.1	0.5	89	CR	Y
17	0.2	4.2	0.16	252	CR	6.4	1.1	0.18	0.28	3.8	0.2	201	CR	Y
18	0.2	3.9	0.3	154	PR									Y
19	0.18	4.36	0.29	177	CR	0.15	4.2	0.3	0.2	3.9	0.16	320	CR	Y
20	0.15	4.1	0.3	176	CR	0.2	3.9	0.26	0.14	4.1	0.2	264	CR	Y
21	0.12	4.1	0.3	168	CR	0.12	4	0.26	0.2	3.9	0.3	168	CR	Y
22	0.1	4.1	0.2	281	CR	0.1	4.2	0.2	0.14	3.9	0.3	187	CR	Y
23	0.2	3.8	0.3	138	CR									Y
24	0.14	4.1	0.33	155	CR	0.1	3.9	0.3	0.11	4.2	0.3	171	CR	Y
25	0.2	3.6	0.16	305	CR	1.2	3	0.2	0.2	4.1	0.3	162	CR	Y
26	2	3.9	0.21	224	PR	3.6	2.2	0.3	3.1	3	0.2	235	NR	Y
27	0.11	4.1	0.16	284	CR	1.2	3.1	0.2	0.11	4.1	0.3	151	CR	Y
28	0.4	3.6	0.24	224	CR	0.2	4.1	0.3	0.1	4	0.2	268	CR	Y
29	1	3.1	0.4	109	PR	1.2	3.5	0.5	0.2	4.2	0.45	97	CR	Y

S.No.	If yes, duration off steroids (in months)	If not, steroid dependent dose (mg/kg/day)	Off CNI (Y/N)	If yes, CNI stopped after how many months	If yes, duration off CNI (in months)	If no or if restarted, CNI dose (mg/kg/day)	CNI drug levels (if done)	First relapse after change in therapy (months)	No. of relapses after change in therapy (in 6 months)	No. of relapses after change in therapy (in 12 months)	S. creat at stopping CNI (if stopped)(in mg/dl)	Treatment failure (Y/N)	If yes, cause of treatment failure
1	10		N			0.04		7	0	1	0.3	N	
2	11		Y	3	9				0	0	0.4	N	
3	12		Y	5	7				0	0	0.5	N	
4	5		Y	5	2	0.04		6	1	1	0.31	N	
5	9		N			0.02		11	0	1	0.2	N	
6	1.5		N			0.04		5	1	1	0.26	N	
7	8		N			0.06			0	0	0.3	N	
8	3		N			0.04		8	0	2	0.4	N	
9	4		N			0.25		6	1		0.3	N	
10	4		Y	3	9			2	1	1	0.45	N	
11	6		N			0.15		7	0	1	0.77	N	
12	6		N			0.07			0	0		N	
13	2		N			0.1		8	0	1	0.45	N	
14	3		Y	4	8			9	0	1	0.4	N	
15		0.09	N			0.07		2	1	2	0.7	Y	COULD NOT WITHDRAW STEROIDS OR CNI
16	2		N			0.06		5	1	2	0.5	N	
17	6		N			0.05		2	1	1	0.5	N	
18	5		N			0.08		5	1		0.4	N	
19	10		N			0.03			0	0	0.6	N	
20	11		N			0.03			0	0	0.3	N	
21	12		Y	10	2				0	0	0.5	N	
22	12		Y	11	1				0	0	0.6	N	
23	3		Y	0	12			1	3		0.7	Y	FREQUENT RELAPSES, >2 IN SIX MONTHS
24	12		Y	9	3				0	0	0.38	N	
25	12		Y	11	1			7	0	1	0.34	N	
26	1		N			0.08		5	1	3	0.2	Y	FREQUENT RELAPSES, >3 IN TWELVE MONTHS
27	3		Y	10	2			8	0	1	0.3	N	
28	10		Y	9	3			4	1	1	0.2	N	
29	12		N			0.05		4	1	2	0.45	N	

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No. BFUHS/2K23p-TH/ 74 Date 01/01/24

The Principal
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Subject:: Approval of Plan of Thesis.

This is to inform you that the Faculty of Medical Sciences in its meeting held on 25.09.2023 has approved the plan of thesis of **Mahak**, a student of your college for **DM Nephrology** along with the names of Supervisor/Co-Supervisor as under:-

Topic Plan of Thesis
To study calcineurin inhibitor sparing strategies in patients with steroid resistant and steroid dependent nephrotic syndrome .

Supervisor
Dr. Vikas Makkar
Prof. & Head
Dept. of Nephrology

Co-Supervisor:-
Dr. Gurdeep Singh Dhooria
Professor
Dept. of Paediatrics

Co-Supervisor:-
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Dept. of Nephrology

The student may submit the thesis in triplicate alongwith the plan in triplicate also, to the Controller of Examinations, BFUHS, Faridkot, at least six months before the commencement of theory examination, if he/she is otherwise eligible to take the examination. He /She may be directed to attach a photocopy of this letter with the forwarding letter while submitting thesis in the University along with the letter of approval of University for change of supervisor/Co-Supervisor, change of topic, if any.

Controller of Examinations

Note :

1. *If any discrepancy found regarding topic of thesis or Supervisor/Co-Supervisor, inform undersigned within one month after issue of this letter.*

TO STUDY CALCINEURIN INHIBITOR SPARING STRATEGIES IN PATIENTS WITH STEROID RESISTANT AND STEROID DEPENDENT NEPHROTIC SYNDROME

THESIS PROPOSAL

FOR APPROVAL OF SUBJECT OF THESIS TO BE SUBMITTED

IN PARTIAL FULFILMENT OF THE REQUIREMENTS

FOR THE DEGREE OF

DOCTORATE OF MEDICINE

IN

NEPHROLOGY

OF THE

BABA FARID UNIVERSITY OF HEALTH SCINECES
FARIDKOT

OCTOBER, 2022

DR. MAHAK

DEPARTMENT OF NEPHROLOGY
DAYANAND MEDICAL COLLEGE AND HOSPITAL
LUDHIANA

ABTRACT OF PLAN OF THESIS

Title	TO STUDY CALCINEURIN INHIBITOR SPARING STRATEGIES IN PATIENTS WITH STEROID RESISTANT AND STEROID DEPENDENT NEPHROTIC SYNDROME
For the degree of	DM Nephrology
Name of the candidate	Dr. Mahak
Supervisor	Dr. Vikas Makkar, Professor & Head, Department of Nephrology, DMCH, Ludhiana
Co-supervisor(s)	Dr. Gurdeep Singh Dhooria, Professor, Department of Pediatrics, DMCH, Ludhiana Dr. Simran Kaur, Associate Professor, Department of Nephrology, DMCH, Ludhiana
Institution	Dayanand Medical College & Hospital, Ludhiana
University	Baba Farid University of Health Sciences, Faridkot

Introduction: Nephrotic Syndrome is one of the most common chronic kidney diseases (CKD) in children . The management of patients with steroid resistance and dependence is challenging. Because of variable response to immunosuppression, therapy- and disease related adverse effects, alternative or additive agents have been sought for maintenance therapy in these patients

Aim of Study: To observe the treatment response to various drugs that can be used as CNI sparing agents for maintaining remission in patients of SRNS and SDNS

Materials & Methods: It would be an observational study, done in the department of nephrology and paediatrics. Patients of steroid resistant (SRNS) and steroid dependent nephrotic syndrome (SDNS; with infrequent relapses on CNI), who have completed atleast one year of therapy with CNI and are in complete or partial remission at the time of enrollment would be included. The effect of various treatment strategies employed as CNI sparing agents would be studied for the incidence of sustained remission , relapses and side effects observed.

Clinical Significance: CNI are effective in maintaining remission in patients with SRNS and SDNS. But significant adverse effects argue for switching to less toxic agents. This study would guide about the effective CNI sparing strategies that can be employed for maintaining remission in these patients.

Keywords: steroid resistant nephrotic syndrome (SRNS), Steroid dependent nephrotic syndrome (SDNS), Calcineurin Inhibitor (CNI), Mycophenolate Mofetil (MMF).

(Signature)	(Signature)	(Signature)	(Signature)	(Signature)
Candidate	Co-supervisor	Co-supervisor	Supervisor	Principal

CERTIFICATE OF FACILITIES AVAILABLE

This is to certify that the facilities for work on the subject of thesis titled **"TO STUDY CALCINEURIN INHIBITOR SPARING STRATEGIES IN PATIENTS WITH STEROID RESISTANT AND STEROID DEPENDENT NEPHROTIC SYNDROME"** exist in the Department of Nephrology, DMC&H, Ludhiana and will be provided to the candidate. We will see that the data being included in the thesis is genuine and is collected by the candidate herself under our supervision and guidance.

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Date: _____

APPROVAL PROFORMA BY
RESEARCH & ETHICAL COMMITTEE
DAYANAND MEDICAL COLLEGE & HOSPITAL, LUDHIANA

Name of candidate	Dr. Mahak
Department	Nephrology
Topic of Thesis	TO STUDY CALCINEURIN INHIBITOR SPARING STRATEGIES IN PATIENTS WITH STEROID RESISTANT AND STEROID DEPENDENT NEPHROTIC SYNDROME
Likely date of appearing in PG examination	May/June, 2024
Date of enrollment	26.04.2022
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Dr. Harpreet Kaur (Professor & Head, Pathology)	Mr. R K Sharma (Legal Expert)
Dr. Jaspal Singh (Professor & Head, Surgery)	Dr. Ramit Mahajan (Associate Professor, Gastroenterology)
Dr. Dinesh Gupta (Professor & Head, Medicine)	Dr. Anu Sharma (Professor, Anatomy)
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Dr. Gurpreet S Wander (Vice Principal)	Dr. Gurpreet S Wander (Vice Principal & Professor & Head, Cardiology)
	Dr. Gagandeep Singh (Secretary, Ethics Committee Professor & Head, Neurology)

Approved: Yes / No

Dr. Sandeep Puri
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Research Committee

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Research Committee

Approved: Yes / No

Prof. Arvind Malhotra
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Ethics Committee

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BABA FARID UNIVERSITY OF HEALTH SCIENCES, FARIDKOT

PROFORMA FOR REGISTRATION OF SUBJECT FOR DISSERTATION

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4	DATE OF ADMISSION OF COURSE	29.04.2022
5	TITLE OF THE TOPIC	TO STUDY CALCINEURIN INHIBITOR SPARING STRATEGIES IN PATIENTS WITH STEROID RESISTANT AND STEROID DEPENDENT NEPHROTIC SYNDROME

6. BRIEF RESUME OF THE INTENDED WORK

6.1 NEED FOR THE STUDY

Nephrotic Syndrome is one of the most common chronic kidney diseases (CKD) in children with an estimated incidence of 2.92 (range 1.15-16.9) per 100,000 children per year.[1] It is characterized by nephrotic range proteinuria; ≥3+ by dipstick, proteinuria ≥ 40 mg/m2/hr (>1000 mg/m2/day), urine protein to creatinine ratio (Up/Uc) ≥2mg/mg; hypoalbuminemia (<3 g/dl); and edema.[2]

Approximately 85% of cases experience complete remission of proteinuria following daily prednisolone/prednisone (PDN) treatment at standard doses.[3] Those who do not achieve remission after 4-6 weeks of therapy with prednisone are presumed to have steroid resistant nephrotic syndrome (SRNS).[4] For those children who respond to steroids, the majority will have one or more relapses and half will have frequently relapsing (≥4 relapses/year) or steroid-dependent (two consecutive relapses during steroid therapy or within 14 days of stopping steroids) nephrotic syndrome (SDNS).[5]

The management of patients with steroid resistance is challenging because of variable response to immunosuppression, therapy- and disease related adverse effects, risk of progression to kidney failure, and likelihood of post-transplant recurrence.[4] With any immunosuppressive strategy, the intent is to induce remission of proteinuria, which is associated with favourable long term outcomes.

Current guidelines recommend calcineurin inhibitors (CNI) as first line therapy for patients with initial or late steroid resistance.5 The optimal duration of CNI therapy varies in different guidelines. KDIGO and IPNA recommend therapy for at least 1 year), with tacrolimus being preferred to cyclosporine. Similarly in patients with SDNS, guidelines recommend the use of steroid sparing agents like CNI, cyclophosphamide, levamisole and mycophenolate mofetil.

There is lack of information regarding the efficacy and safety of available pharmacological options during long term follow up to maintain remission. Since prolonged use of high-dose CNI can result in undesirable side effects (i.e. nephrotoxicity), alternative or additive

drugs have been sought; like iv or oral cyclophosphamide, rituximab, MMF, ACE/ARBs and monoclonal antibodies such as ofatumumab.

Mycophenolate mofetil (MMF), the pro drug of mycophenolic acid (MPA), may be a promising therapeutic option without nephrotoxicity in maintaining remission in patients with SRNS and SDNS. Multiple case series show that therapy with MMF is effective and safe in reducing the frequency of relapses and cumulative corticosteroid requirement in patients with steroid-dependent nephrotic syndrome.[6] and SRNS.

The aim of this study is to observe the various CNI sparing treatment strategies available to maintain sustained remission in patients with SRNS and SDNS.

6.2 REVIEW OF LITERATURE

The management of patients with SRNS is challenging. Clinicians are often faced with therapeutic dilemmas and challenges while treating children with frequently relapsing steroid-dependent nephrotic syndrome (SDNS) and steroid-resistant nephrotic syndrome (SRNS). SRNS is associated with unsatisfactory patient-reported quality of life, morbidity due to infectious and non infectious illnesses, and side effects of therapy.[4]

In the absence of studies to determine the appropriate duration of CNI therapy in patients with complete or partial remission, KDIGO and IPNA recommend therapy for at least 1 year, while ISPN advises therapy for 2 years or longer. Therapy should be monitored closely for adverse effects.

Calcineurin inhibitors (CNIs) inhibit T-cell activation and may be exerting their effect in nephrotic syndrome through this mechanism. Cyclosporine and tacrolimus are the two CNIs commonly used. Most centres prefer tacrolimus over cyclosporine because of its better efficacy and less systemic side effects, particularly nephrotoxicity, except in children who are unable to swallow tablets and in patients with seizures or at risk of diabetes. Doses of CNIs are titrated to achieve recommended trough levels, keeping in mind interaction with other medications.

Most patients who respond to CNI do so within the first 6 months of treatment.[7] About 10-25% of patients receiving prolonged CNI treatment are at risk of nephrotoxicity.[8] Risk factors for nephrotoxicity include presence of initial resistance, dose of CNI used, duration of heavy proteinuria, and hypertension during therapy. Adverse effects associated with therapy, including glucose intolerance and acute and chronic nephrotoxicity argue for switching to less toxic agents to maintain CNI-induced remission. In patients with CNI induced nephrotoxicity, available options are mycophenolate mofetil or iv rituximab. Consensus guidelines on management of SRNS 5 suggest treatment with iv cyclophosphamide in patients with non-availability of

CNI, either due to its cost or adverse effects. Another option is to discontinue therapy if patient has been in sustained remission.[5]

Similarly, all guidelines recommend the use of steroid sparing agents in patients with frequent relapses or steroid dependence. However, few randomized trials have compared various interventions for their relative efficacy and safety. In the absence of a strong evidence base, the choice of steroid sparing therapy is based on patient age, presence of steroid toxicity, adherence, risk of adverse effects, patient preference, and availability of medications.

The lack of renal, hemodynamic, and metabolic adverse effects with mycophenolate mofetil (MMF), the prodrug of mycophenolic acid, is an important consideration in preferring this agent over CNIs when treating difficult cases of nephrotic syndrome.[9] MMF suppresses the immune system through inhibition of B- and T- lymphocyte proliferation. It is gaining popularity as a treatment of NS due to its favourable side effect profile compared to CNIs, specifically its lack of renal toxicity. There is a poor correlation between MMF dose and mycophenolic acid (MPA) exposure and significant inter- and intra-patient variability in drug pharmacokinetics. Case series suggest that therapy with MMF and corticosteroids might induce remission in such patients.[10]

In a National Institutes of Health–funded, randomized multicentre trial, the efficacy of a 12-month course of cyclosporine was compared to a combination of MMF and oral dexamethasone in patients with steroid-resistant focal segmental glomerulosclerosis (FSGS). Partial or complete remission occurred in 46% of cyclosporine-treated patients and 33% of MMF+ dexamethasone treated patients after 12 months. They did not find a significant difference in rates of proteinuria remission in the two groups.[11]

Other reports suggest that add on therapy with MMF enabled remission in 20% to 50% patients refractory to treatment with CNIs and corticosteroids. S Hibino et al performed a retrospective study to assess the 3 year outcome of a unified protocol for childhood idiopathic nephrotic syndrome. They found that treatment with CsA and

combination of CsA plus MMF was useful for SDNS and for remission induction in SRNS.

In a retrospective study on 23 patients by Gellerman et al, in patients of SRNS who attained complete remission with cyclosporine therapy, it was attempted to wean CsA and to maintain remission with MMF monotherapy. MMF was started at a low dose followed by a reduction of CsA dose stepwise. They found that therapy with cyclosporine for mean duration of 1.7 years could be successfully switched to MMF in 79% cases.[12]

Li Z et al recruited 24 children (18 boys and six girls) with steroid-resistant idiopathic nephrotic syndrome (SRINS) who were <2 years. All patients were administered prednisone 2 mg/kg per day prior to mycophenolate mofetil (MMF). By the end of the eighth week, MMF was initiated at 25-30 mg/kg daily for 6- 12 months. Prednisone dose was reduced stepwise. With MMF treatment, the levels of urinary protein and serum cholesterol decreased and that of serum albumin increased in a time-dependant manner. They demonstrated that MMF could reduce proteinuria in SRNS children <2 years and suggested that MMF therapy might be an effective strategy for treating SRNS in children <2 years.[13]

. Experience from uncontrolled studies indicates that MMF is perhaps more efficacious in young children[14] and superior to levamisole in steroid dependence. Guidelines from the German Society of Paediatric Nephrology (GPN) limit the choice of steroid sparing medications to chiefly CNI or MMF.[15]

Retrospective case series show that MMF enables CNI- and corticosteroid-free remission in some patients with CNI dependence and/or toxicity. In a prospective study on the use of mycophenolate mofetil in children with cyclosporine-dependent nephrotic syndrome, Fujinaga S et al analyzed 12 children with idiopathic steroid-dependent NS requiring long-term CsA therapy with MMF for at least 6 months. Treatment with MMF resulted in CsA and/or prednisolone (PSL) sparing, with a reduction in mean CsA dose from 3.5±1.3 to 1.5±2.4 mg/kg/day ($p<0.01$), and mean PSL dose from 0.29±0.16 to 0.21±0.11

mg/kg/day ($p<0.05$). They, thus, demonstrated that MMF therapy results in significant CsA and/or steroid sparing and reduction in relapse rates in children with CsA-dependent NS.[16]

A combination of CNI and MMF was shown to induce remission in 67.3% of 49 patients included in four case series of patients with CNI-refractory illness , without significant adverse effects.[17]

The efficacy of MMF monotherapy in achieving initial remission is currently unknown since controlled studies have not been performed.

Rituximab, a chimeric anti CD 20 monoclonal antibody, has been successfully used in patients with frequently relapsing and steroid dependent nephrotic syndrome.

Ravani et al presented long-term follow-up data from children with Idiopathic nephrotic syndrome dependent on both prednisone and calcineurin-inhibitors for at least one year and in remission for at least six months. Consecutive children with these characteristics were treated with one or more rituximab infusions (up to 5). They showed that remission can be achieved for 6 months in half of these children, with longer remission times following repeated treatments, greater response rates for single oral agent-free remission and better outcomes in older children.[18]

Gulati A et al in their study showed that therapy with rituximab was safe and effective in inducing and maintaining remission in a significant proportion of patients with difficult SRNS and SDNS.[19]

A recent meta-analysis on the Efficacy and Safety of Rituximab for Childhood Steroid-Dependent Nephrotic Syndrome included six RCTs and one retrospective comparative control study, involving a total of 446 patients, including 231 in the rituximab group and 215 in the control group. Data analysis results showed that compared with the control group, the RTX treatment group could significantly improve the complete remission rate of children with SDNS. In addition, the RTX treatment group was better than the control group in improving the level of serum albumin and estimated glomerular filtration rate, and the difference was statistically significant.[20]

Furthermore, data on children of Indian ascent treated with iv cyclophosphamide have shown that a small subset of patients do achieve complete remission, either when iv cyclophosphamide was used as first line therapy or after failure of a trial of high dose iv steroids.[21]

Another study in North American children with difficult to treat SRNS demonstrated that iv cyclophosphamide could be useful strategy to induce remission in a subset of children with minimal change disease on biopsy.[22]

Further studies are needed to test the efficacy of MMF, cyclophosphamide and rituximab in inducing remission and, as part of a combination therapy, achieving sustained remission in patients with SRNS and SDNS.

6.3 AIMS AND OBJECTIVES OF THE STUDY

AIM: To study the various CNI sparing treatment strategies in SRNS AND SDNS after one year of therapy with CNI

PRIMARY OBJECTIVE: To observe the incidence of sustained remission after change in therapy

SECONDARY OBJECTIVES:

1. To observe the incidence of relapses prior to and after the change in therapy
2. To observe the side effect profile prior to and after change in therapy including renal dysfunction (GFR)

7. MATERIALS AND METHODS

7.1 SOURCE OF DATA:

Inpatients and outpatients in Department of Nephrology and Paediatrics, DMCH, Ludhiana

7.2 METHOD OF COLLECTION OF DATA :

Patients of SRNS and SDNS who have already received atleast one year of therapy with any of the CNI (cyclosporine or tacrolimus) would be enrolled. Baseline demographic values, laboratory values including GFR, side effect profile would be recorded.

Various treatment strategies that can be employed are:

1. Rituximab, given at a dose of 375 mg/m2 2 doses , 1 week apart while monitoring CD 19 levels (target reduction to <1%)
2. Intravenous cyclophosphamide 500-750 mg/m2 ; every month for 6 months
3. Oral cyclophosphamide (2-2.5 mg/kg) given for 8-12 weeks ; not to exceed 168 mg/kg cumulative dose
4. Continuing with low dose steroids with/without ACE-I/ARBs
5. Adding mycophenolate mofetil (MMF) with low dose CNI

The incidence of sustained remission, number of relapses, any infection, side effects , GFR and number of antihypertensives used would be recorded initially monthly and then after every 3 months .

Definitions:

1. Nephrotic syndrome would be defined as: nephrotic range proteinuria; ≥3+ by dipstick, proteinuria ≥ 40 mg/m2/hr (>1000 mg/m2/day), urine protein to creatinine ratio (Up/Uc) ≥2mg/mg; hypoalbuminemia (<3 g/dl); and edema.

2. Steroid resistant nephrotic syndrome : Those who do not achieve remission after 4-6 weeks of therapy with prednisone
3. Steroid-dependent nephrotic syndrome: patients who have two consecutive relapses during steroid therapy or within 14 days of stopping steroids
4. Frequent relapse would be defined as: ≥ 2 relapses in first 6 months from disease onset; ≥ 4 relapses in 12 months period

Relapse would be defined as: urine protein ≥3+ (urine protein/ urine creatinine ratio >2mg/mg) for 3 consecutive early morning specimens, having been in remission previously.

Relapse would be treated as per the nature of disease as:
In SDNS: daily therapy with prednisone at 60 mg/m2 or 2mg/kg untilnil/trace proteins , followed by 40 mg/m2 or 1.5 mg/kg on alternate days for 4 weeks.
In SRNS: Tacrolimus dose would be increased to maintain the trough level between 4-8 ng/ml. Oral prednisone would be administered until remission is achieved or for a maximum period of 4 weeks or 1.5 mg/kg on alternate days for 4 weeks.
In all the patients, renal biopsy would be done if clinically indicated.

INCLUSION CRITERIA

Patients of steroid resistant (SRNS) and steroid dependent nephrotic syndrome (SDNS) who are between 3 months to 18 years of age who have received atleast one year of therapy with either of the calcineurin inhibitors (tacroilimus or cyclosporine) and are in complete or partial remission at the time of initiation of study.

EXCLUSION CRITERIA

1. Patients with secondary form of nephrotic syndrome , or as part of systemic diseases.

2. Patients who have taken CNI for less than 1 year
3. Patients who have frequent relapses on CNI (≥ 3 relapses)
4. Patients with congenital nephrotic syndrome
5. Patients who do not give consent for the study

Study Design –

Observational study.

Duration Of Study/No. of Cases –

December 2022 to November 2023 (minimum 6 months period for each patient)

OUTCOME

The incidence of sustained remission, No. of Relapses and side effect profile after change in therapy.

STATISTICAL ANALYSIS

Data will be described in terms of range; mean ±standard deviation (± SD), median, frequencies (number of cases) and relative frequencies (percentages) as appropriate. All statistical calculations will be done using (Statistical Package for the Social Science)SPSS 21version (SPSS Inc., Chicago, IL, USA) statistical program for Microsoft Windows.

7.3 DOES THE STUDY REQUIRE ANY INVESTIGATIONS OR INTERVENTIONS TO BE CONDUCTED ON PATIENTS OR OTHER HUMANS OR ANIMALS? IF SO, PLEASE DESCRIBE BRIEFLY.

CNI Level, Renal biopsy if clinically indicated.

7.4 HAS ETHICAL CLEARANCE BEEN OBTAINED FROM YOUR INSTITUTION IN CASE OF 7.3?

Yes, Attached herewith.

8. LIST OF REFERENCES

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PROFORMA

Name: Age:

Sex: Urban/Rural:

MRD No

Diagnosis SDNS/SRNS

Steroid Resistant: Late / Early

Date of birth:

Age Of Onset (Year / Month):

Hypertension at onset:

Haematuria at onset:

If SDNS: Dependent Dose (Mg/Kg/Day):

No of relapses on CNI therapy :

Prior Immunosuppressives:

1)Levamisole: Y/N Duration:

2)Cyclophosphamide :Y/N Duration:

3) Mycophenolate: Y/N Duration

Status Of Patient At Start Of study (i.e. after one year of therapy with CNI) : CR/ PR

Features of steroid Toxicty: Cataract/ Cushingoid / Short Stature/ Overweight/ Obesity/ Behavior abnormality

Renal Biopsy Findings(Year):

SIGNATURE OF CANDIDATE SIGNATURE OF SUPERVISOR

Side effects of CNI: (Tick)

Hypertension	Reversible Nephropathy
Hyperkalemia	Hypertrichosis
Seizure	Gingival hyperplasia
Increase Blood Sugar	Acne
Increase OT/ PT	Skin infections
Diarrhea	Major infection (Requiring hospitaliza-tion)Diagnosis
UTI	Others

Date Of Start Of study

Age YRS	Weight KG	Height CM	BMI	Sr Creati-nine	Sr Albumin
Z SCORE					

Baseline Investigation: CBC RFT,

OT,PT S.Albumin,

S.Cholesterol Urine PCR

Tac levels

Strategy implied:

Dose Given: _____

Associated Medications:

SIGNATURE OF CANDIDATE SIGNATURE OF SUPERVISOR

Follow up:

Date / Months	Date of enrolment	Dose of CNI being used ME	Drug levels	Prednisolone (alternate/ daily)	Drug added	s. al-al-bumin	s. creatinine	Edema	Response CR/ PR/ NR)	Remission/ relapse	Any other medication being used	Specific drug related side effects	CD 19 levels (if rituximab is used)

SIGNATURE OF CANDIDATE SIGNATURE OF SUPERVISOR

Results

Status Of Patients At 6 Months Post change in therapy: CR / PR/ NR

Off Steroids: Y/N_____ (IN SDNS)

If Yes, Duration Off Steroids: _____

If No, Steroid Dependant Dose: _____

Off CNI : Y/N_____ (IN SDNS/SRNS)

If Yes, Duration Off CNI: _____

If No, CNI Dose: _____

CNI drug levels (if reqd)

First Relapse (Months after changing therapy):

No. Of Relapses After changing therapy (In 6- 12 Months Period):

Sr Creatinine at stopping CNI:

Treatment failure: **Y/N**

CAUSE OF TREATMENT FAILURE

Recurrence of SR (no response after relapse, after 4 weeks full dose steroids) y/n

FR ≥ 2 relapses in six months/ ≥ 3 in 12 months

y/n

Irreversible increase in Sr Creatinine (< 50 ml/min/ 1.73m2)
y/n

Shifting of patients with steroid dependent to steroid resistant state

Withdrawal of consent

SIGNATURE OF CANDIDATE SIGNATURE OF SUPERVISOR

INFORMED PATIENT CONSENT

Date: _____

I……..D/o, W/o, S/o……. ……am willing to participate in the study **"TO STUDY CALCINEURIN INHIBITOR SPARING STRATEGIES IN PATIENTS WITH STEROID RESISTANT AND STEROID DEPENDENT NEPHROTIC SYNDROME"** The investigation has been explained to me in local vernacular language. I have fully understood that no extra expenditure in any form, merely for the purpose of thesis is being carried out. I am free to opt out the study any time without assigning any reason.

…….	…….
Signature of the Witness	Signature of the patient / Legal representative
Name:	Name:

Signature of Candidate **Signature of Supervisor**

सूचित रोगी की सहमति

दिनांक: _____

मैं …….. डी/ओ, डब्ल्यू/ओ, एस/ओ ……। …… मैं **"TO STUDY CALCINEURIN INHIBITOR SPARING STRATEGIES IN PATIENTS WITH STEROID RESISTANT AND STEROID DEPENDENT NEPHROTIC SYNDROME"** अध्ययन में भाग लेने के लिए तैयार हूं। जांच के बारे में मुझे स्थानीय स्थानीय भाषा में समझाया गया है। मैं पूरी तरह से समझ गया हूं कि किसी भी रूप में कोई अतिरिक्त खर्च नहीं किया जा रहा है, केवल थीसिस के उद्देश्य से किया जा रहा है। मैं बिना कोई कारण बताए किसी भी समय अध्ययन से बाहर निकलने के लिए स्वतंत्र हूं।

…… …… ……

गवाह के हस्ताक्षर रोगी के हस्ताक्षर /कानूनी प्रतिनिधि

नाम नाम:

उम्मीदवार के हस्ताक्षर पर्यवेक्षक के हस्ताक्षर

ਸੂਚਿਤ ਮਰੀਜ਼ ਦੀ ਸਹਿਮਤੀ

ਤਾਰੀਖ਼: _____

I……..D/o, W/o, S/o…… ਮੈਂ **“TO STUDY CALCINEURIN INHIBITOR SPARING STRATEGIES IN PATIENTS WITH STEROID RESISTANT AND STEROID DEPENDENT NEPHROTIC SYNDROME”** ਅਧਿਐਨ ਵਿੱਚ ਹਿੱਸਾ ਲੈਣ ਲਈ ਤਿਆਰ ਹਾਂ। ਜਾਂਚ ਮੈਨੂੰ ਸਥਾਨਕ ਭਾਸ਼ਾ ਵਿੱਚ ਸਮਝਾਈ ਗਈ ਹੈ। ਮੈਂ ਪੂਰੀ ਤਰ੍ਹਾਂ ਸਮਝ ਗਿਆ ਹਾਂ ਕਿ ਕਿਸੇ ਵੀ ਰੂਪ ਵਿੱਚ ਕੋਈ ਵਾਧੂ ਖਰਚਾ ਨਹੀਂ, ਸਿਰਫ਼ ਥੀਸਿਸ ਦੇ ਉਦੇਸ਼ ਲਈ ਕੀਤਾ ਜਾ ਰਿਹਾ ਹੈ। ਮੈਂ ਬਿਨਾਂ ਕਿਸੇ ਕਾਰਨ ਦੱਸੇ ਕਿਸੇ ਵੀ ਸਮੇਂ ਅਧਿਐਨ ਤੋਂ ਹਟਣ ਲਈ ਸੁਤੰਤਰ ਹਾਂ।

…… ……

ਮਰੀਜ਼ ਦੇ ਗਵਾਹ ਦੇ ਦਸਤਖਤ /ਕਾਨੂੰਨੀ ਪ੍ਰਤੀਨਿਧੀ

ਨਾਮ: ਨਾਮ:

ਸੁਪਰਵਾਈਜ਼ਰ ਦੇ ਉਮੀਦਵਾਰ ਦੇ ਦਸਤਖਤ